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PROCEEDINGS
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VOL. XXX

SECTION - B

PART III

A STUDY ON THE AQUATIC PLANT COMMUNITY AT GORAKHPUR
IN RELATION TO THE pH OF THE MEDIUM

By

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[Received on 2nd June, 1959]

INTRODUCTION

The study of pH in relation to plant communities has attracted attention of a number of workers. But it seems that not enough attention has been bestowed on the problem of tolerance of pH of the medium by aquatic plants. Seasonal variations of pH depend on a number of environmental factors which play a major part in determining the character of any plant community; water happens to be one of the most important of such factors. Even in the case of rooted aquatic plants, morphology and physiology are to a great extent affected by the surrounding medium. Such being the case, the physical and chemical characteristics of the water are likely to influence the organisation of the plant body. The contact surfaces of the entire plant in such cases function in absorption, the roots in this respect playing a comparatively minor role.

So far as the land plants are concerned data have been reported which relate to the soil pH as a conditioning factor in the absorption of nutritive substances from the soil. The pH is usually determined by the reaction of an aqueous extract of the soil, and for this purpose different ratios of soil and water have been employed for preparing extracts, as for example 1:3 (Misra, 1946) 1:5 (Puri, 1950), 1:10 (Srivastava & Tandon, 1951). Puri (1949) has found that values are not significantly altered even though the ratios are varied from 1:5 to 1:25. In the case of aquatic plants, this difficulty does not present itself, because the environmental water more suitably replaces the soil extract for pH determination.

The authors have conducted a survey of the vegetation of a number of ponds near about Gorakhpur (25°5' N and 83°8' E) with a view to determining its relation to the changes in the pH of the surrounding medium consequent upon the progressive drying up of the water from the time of the rains to the advent of the comparatively dry winter season.

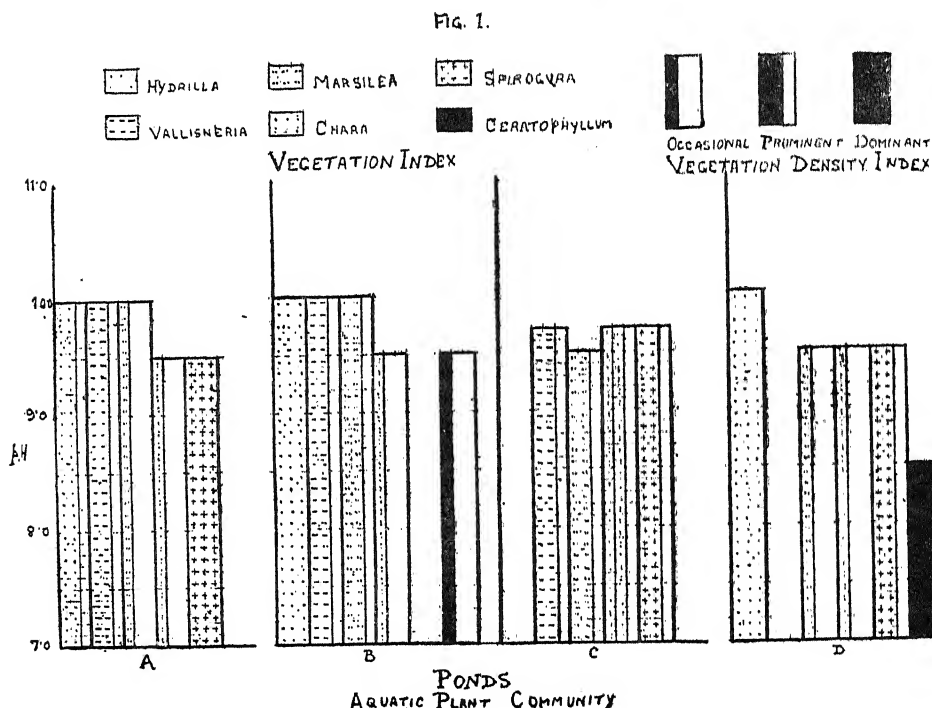
METHOD

Samples of surface water were collected and their pH determined at various times. This operation was continued till the ponds more or less dried up. The pH of each sample of water was determined right at the place of collection for it seemed likely that changes in the pH might occur during the lapse of time which would be inevitable if the samples were collected and brought to the laboratory for investigations. The pH was determined by the colorimetric method with the help of B. D. H Universal Indicator and Indicator Test Papers, and also other B. D. H indicators, the comparison being finalised by the All-Purpose Lovibond Comparator.

OBSERVATIONS

Field observations were made at different times starting from the advent of the rains to the comparatively dry weather when the ponds more or less dried up. About 50 ponds were brought within the range of this study.

The observations thus made have been summarised in the case of four ponds only, viz., A, B, C and D situated in different areas in this locality, and presented graphically in Fig. 1. It represents data concerning the population of an aquatic



plant community consisting of *Hydrilla verticillata*, *Vallisneria spiralis*, *Marsilea quadrifolia*, *Chara* sp., *Spirogyra* sp. and *Ceratophyllum demersum* in which the density of the individual species at a particular pH is represented by the thickness of the perpendicular axis in the order in which the species have been named.

It was observed that soon after the rainy season the pH value seldom fell below 7 and varied between 7 and 7.5, indicating a slight alkaline reaction. The four

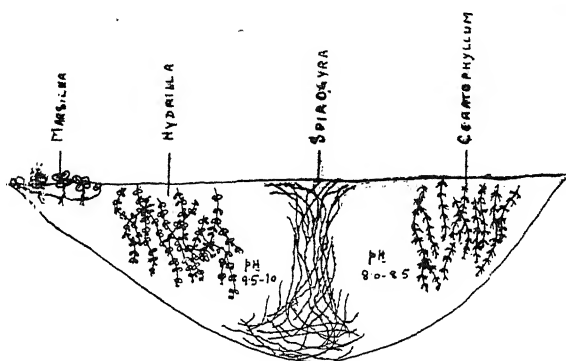
ponds mentioned above have been described in detail in the following paragraphs.

Pond A. This is situated near the Domingarh railway bridge on one side of North Road. Water was 5-7 feet deep in the rainy season. By January the water level went down to about 1-2 feet or even less. A sudden rise of pH was noted at this time when it had luxuriant growth of the aquatic plant community indicated above, with the exception of *Ceratophyllum demersum*. *Vallisneria spiralis* had complete red-brown leaves. The pH value ranged from 9.5 to 10, during the comparatively dry period of the year beginning with the onset of winter.

Pond B. The pond B is situated on the left side of Domingarh Road, a little ahead of the pond D described below. It occupies a large area during the rainy season, though it is not as deep as pond A. In February the water was 1 foot to 6 inches deep or even less. At this time it had a luxuriant vegetation consisting of *Hydrilla verticillata*, *Vallisneria spiralis* and *Marsilea quadrifolia*. Other plants of the community indicated above were less common. The pH value ranged from 9.5 to 10 in comparatively dry periods when water level was low.

Pond C. The pond C is situated near Turra Nulla of Tinkonia Forest Range in the north of Gorakhpur. A great variation of pH at different places in the same pond was found. In areas indicating low pH values the vegetation was very much sparse, whereas in areas with high pH, the water had luxuriant plant growth. In addition to the plants mentioned above, the community also contained *Potamogeton crispus*. The pH value ranged from 9.5 to 9.7 in dry winter months of the year.

Pond D. The pond is situated at the end of the road leading to Domingarh. This pond indicated a pH value of 7.5 on the 11th January, 1959, but when examined on the 12th February, 1959, it indicated a peculiar variation of pH in different regions, the kind and density of the aquatic vegetation showing some kind of correlation as indicated above with the prevailing pH at a particular region. The water at this time was 8 to 15 inches deep, and the pond was some what divided into two portions by an abundant vertical growth of *Spirogyra* sp., in the middle of the water. Its inter-twining filaments formed a closely woven partition sinking down from the surface to the bottom due to the formation of zygospores as shown in Fig. 2. In one of the two portions, there was a dense growth of *Cerato-*



A DIAGRAMATIC BISECT OF POND-D.
FIG. 2.

phyllum demersum leading to the practical exclusion of all other vegetation, and the pH in this region ranged from 8 to 8.5, increasing towards the other portion where there was *Hydrilla verticillata*-*Marsilea quadrifolia* association. The growth of *Hydrilla verticillata* was specially luxuriant and the pH in this region ranged from 9.5 to 10 in comparatively dry conditions.

DISCUSSION

Two significant points seem to emerge from this study, viz., that the vegetation becomes progressively luxuriant as the water in the ponds recedes, and with this luxuriance of vegetation the pH value of the water increases.

A difference appears to exist in the pH values of water in a pond and that of its bed. It would therefore be reasonable to infer that the pH value of the surrounding water is a factor which exerts an appreciable influence on the growth of a plant community. Moreover during the rains the water, which accumulates in the pond, brings along with it dissolved substances which might not be present in the soil at the particular point at which the pond is situated. The vegetation of the pond would therefore seem to depend rather on the quality of its water than that of the soil making up the bed. It would therefore follow that the pH value of the soil at the bed would probably influence to some extent the formation of a plant community in the pond only by bringing about changes in the pH value of the pond water. Srivastava and Tandon (1951), while studying the autecology of *Tropha bispinosa* did not take into consideration the pH values of the water on which the plants were afloat; the range of pH values they determined was that of the pond-bed soil, viz., 6.2 to 7.5, the later value indicating a slight alkaline reaction. Kachroo (1956) working on the pond-bed soils grouped ponds in accordance with the pH values of pond-bed soils. The minimum and maximum pH values which he could determine from two pond beds were 6.0 to 6.2 and 7.1 to 7.5 respectively. Of the seven ponds studied by him, four were grouped as showing acid reaction, two as showing reactions varying from acidity to alkalinity, and only one as showing definitely alkaline reaction. Srivastava (1950) studying the pH tolerance of an aquatic plant community consisting of *Hydrilla*, *Vallisneria*, and *Potamogeton* at Allahabad reported pH values of the soil supporting the plants as ranging from 7.2 to 7.6. The observations reported by these authors were all based on the pH determinations of the pond-bed soil, and therefore the values recorded seem to be comparatively low. In the present observations recorded at Gorakhpur and presented in this paper the pH values were determined from water samples and these indicated a persistently alkaline reaction. It will be interesting at this place to refer to the work of Mitra (1955) on the autecology of *Limnanthemum* sp., in which she recorded pH values ranging from 7.3 to 9.2; these observations were based on the pH value determinations of water rather than of the soil to which the plant was rooted.

The pH of the soil is not a constant and characteristic value. In the non-saline alkali soils the pH value ordinarily ranges from 8.5 to 10. The exchangeable sodium hydrolyses in part to sodium hydroxide, which therefore reacts with carbon dioxide to produce a mixture of sodium bicarbonate and sodium carbonate (Black, 1957). It may be visualised that the exchangeable sodium in water, to a somewhat limited extent, is held by the colloidal complex suspended in the water. In the observations reported in this paper, the high pH value was almost always associated with vigorous and luxuriant growth of vegetation, which possibly produced an excessive amount of carbon dioxide forming carbonic acid in water. This would probably bring about a release of sodium ions which hydrolysing to sodium hydroxide might tend to make the pond water alkaline and thus enhance its pH value. This reaction being reversible, the resultant environmental conditions in respect of pH value did not become such as to affect the growing vegetation in an adverse manner.

Black (1957) states that as the exchangeable sodium percentage increases, there is a concomitant increase in the soil pH.

SUMMARY

Observations have been made as regards the relation of pH of water to an aquatic plant community consisting of *Hydrilla verticillata*, *Vallisneria spiralis*, *Marsilea quadrifolia*, *Chara* sp. *Spirogyra* sp. and *Ceratophyllum demersum*. High pH values were recorded from the pond water at the time when the ponds were more or less dried up and the vegetation flourished well. This has been summarised for four ponds graphically to show the density of population of individual plants forming this aquatic plant community. The pH value was almost always above 7, thereby showing the plant community's preference towards alkalinity. The pH values were determined straight from pond water, and not from the soil samples forming the bed which supports this community. A considerable variation in the pH values has been observed by the authors. It appears that surrounding water exerts an appreciable influence on the formation of plant community in the pond.

The correlation between the high pH values of water and the luxuriant vegetation has been discussed.

ACKNOWLEDGEMENT

One of the authors (D. N. S.) is thankful to Prof. M. O. Varkey, Principal, St. Andrew's College, for providing research facilities, and also to his colleague Prof. G. C. Srivastava for help in various ways.

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STUDIES ON SOME CESTODE PARASITES

IV. ON FOUR NEW SPECIES INCLUDING A NEW GENUS BELONGING TO THE FAMILY HYMENOLEPIDIDAE*

By

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[Received on 13th April, 1959]

During the years 1953-56, the author collected a number of cestodes from different avian hosts. Part 4 of the series contains descriptions of 4 new cestodes, one representing a new genus from different places in Uttar Pradesh.

Neoligorchis alternatus, n. g., n. sp.

Three complete worms and a few strobilae were collected from the intestine of *Rostratula bengalensis* Linn. shot in the outskirts of Bareilly district.

Length of the specimens 20-25 mm., maximum width 1.0 mm. External segmentation faintly marked, specially in the middle region of the strobila. Scolex unarmed, much broader than long 0.45-0.55 mm. \times 0.80-0.88 mm. Suckers rounded, diameter 0.52-0.54 mm. Genital pores alternate irregularly located approximately at anterior third of the lateral margin of the segments. Genital cloaca small and poorly developed. Genital ducts pass between poral longitudinal excretory vessels. Cirrus sac simple (0.18-0.21 mm. \times 0.05 mm), extends to and in some segments well past poral longitudinal excretory vessels. Cirrus simple and unarmed. Ductus ejaculatorius coiled. External vesicula seminalis simple but prominent. An internal one is not observed. Testes spherical (average diameter 0.06 mm.) 5-6 in number situated dorsal, anterior and lateral to ovary on the aporal side, partially overlapping it and within longitudinal excretory vessels. Ovary transversely elongated in the middle of the segment, slightly poral in position, 0.22 mm. across. Vagina narrow and posterior to cirrus sac. Receptaculum seminis absent. Vitelline gland nearly spherical, maximum diameter 0.09 mm. lateral to ovary on the aporal side and quite often overlapped by it. Shell gland not observed. Uterus an irregularly lobed sac occupying the entire segment. Eggs elongated, egg shell tapering at each pole with longitudinal striations. Eggs and onchospheres 0.088 mm. \times 0.048 mm. and 0.047 mm. \times 0.032 mm. respectively. Embryonic hook 0.012-0.013 mm. long.

From the characters given above, it is obvious that the present form must belong to the family Hymenolepididae Fuhrmann, 1907 and to the subfamily Hymenolepidinae Perrier, 1897. The form greatly resembles *Drepanidotaenia* Railliet, 1892; *Echinocotyle* Blanchard, 1891; *Hymenosimbria* Skrjabin, 1914; *Chitinoilepis* Baylis, 1926 and *Pseudoligorchis* Johri, 1934. It can, however, be distinguished from *Drepanidotaenia* by the absence of a rostellum, disposition of the testes (testes poral arranged in a transverse row) and the antiporal position of the ovary (in the present form it is slightly poral); from *Echinocotyle* and *Hymenosimbria*

* Part of thesis approved by the University of Lucknow for the award of the Ph. D. degree.

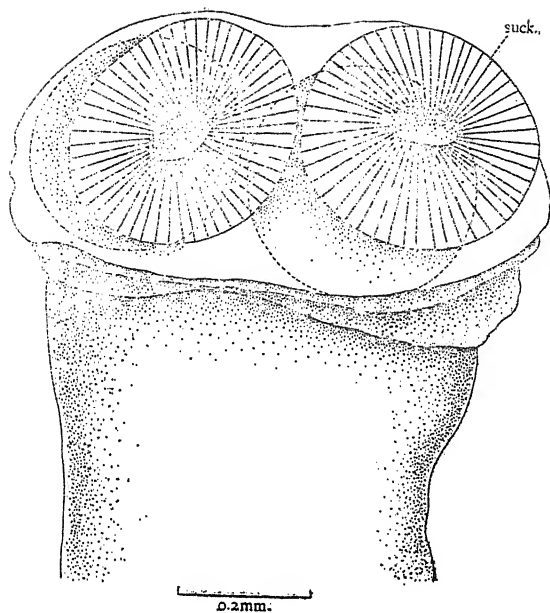


Fig. 1. Scolex of *Neoligorchis alternatus* n. g., n. sp.

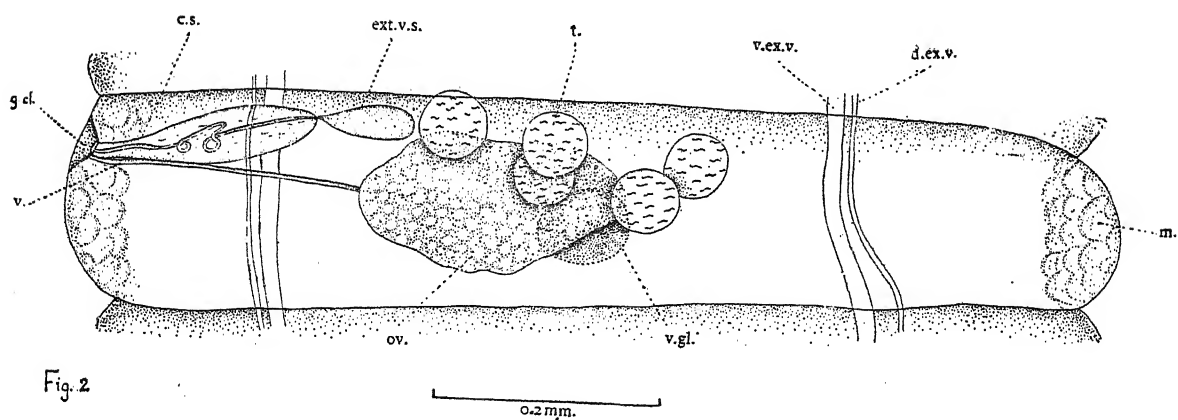


Fig. 2

Mature segment of *Neoligorchis alternatus*

by the absence of a rostellum and a sacculus accessorius and by the number of the longitudinal excretory vessels which are more than two in *Hymenifimbria*. From *Chitinolepis* and *Pseudoligorchis* which also possess an unarmed rostellum and are recorded from mammals, the present form can be separated by the course of the genital ducts, disposition of testes (testes never anterior to ovary) and by the nature of the egg shell which is very thick in *Chitinolepis*.

Neoligorchis n. g.

Generic diagnosis: *Hymenolepididae* : *Hymenolepidinae* :—Scolex unarmed without any rostellum or armature. Genital pores alternating irregularly. Genital ducts pass between poral longitudinal excretory vessels. Testes few, anterior and lateral aporal to ovary. External vesicula seminalis present. Ovary slightly poral. Vitelline gland lateral (aporal) to ovary. Uterus an irregularly lobed sac. Type species: *N. alternatus*.

Hymenolepis ciconia, n. sp.

Two mature worms were collected from the intestine of a white stork, *Ciconia ciconia* Linn. from Bareilly. Scolex was available in only one of the worms.

Length of the worms 30 mm. and 33 mm., maximum width 0.4 mm. attained in fully grown gravid segments. External segmentation prominent, segments typically trapezoidal with serrated margins. Scolex globular 0.19 mm. \times 0.21 mm. Rostellum (0.07 mm. \times 0.05 mm.) well developed armed with a single row of 8 hooks, 0.020-0.022 mm. in length, the blade being smaller about 1/5th the length of the handle. The guard of the hook is blunt and very powerful. Rostellar sac 0.13 mm. \times 0.045 mm. extending almost to the lower level of the suckers. Suckers thick and muscular, average diameter 0.075 mm. Neck short 0.33 mm. long. Segments all broader than long. Immature 0.10 mm. \times 0.22 mm., mature 0.20 mm. \times 0.50 mm. and gravid ones 0.30 mm. \times 0.80 mm. last few segments tend to be rectangular 0.40 mm. \times 0.65 mm. Genital pores unilateral near the middle of the lateral margin of the segments. Genital coeca absent. Dorsal and ventral longitudinal excretory vessels 0.01 mm. and 0.025 mm. in diameter respectively. Cirrus sac (0.17-0.21 mm. \times 0.04-0.06 mm.) obliquely dorsal to and much beyond poral longitudinal excretory vessels and about $\frac{1}{2}$ across the segment. Cirrus simple and unarmed. External vesicula seminalis oval. Testes usually 3 (diameter 0.075-0.10 mm.), two of them aporally one behind the other and the third poral adjacent to posterior aporal testis. Arrangement of testes triangular. Sements, with four testes have two porals and two aporals; with two testes have one poral and the other aporal, the anterior aporal being absent. Ovary bilobed, the two lobes forming the two wings of a characteristic V-shaped structure, each 0.13 mm. long. Maximum width of the ovary 0.13 mm. Vagina posterior to cirrus sac. Receptaculum seminis oval 0.08 mm. in diameter and prominent in gravid segments. Vitelline gland oval posterior to ovary. Uterus saccular within longitudinal excretory vessels. Eggs with double coverings, outer 0.06 mm., inner 0.048 mm. in diameter. Embryonic hooks 0.018 mm. long.

The present form resembles *H. linea* (Goeze, 1782) Wolfhugel, 1899 and *H. longi* Oswald, 1951 in the number and size of the rostellar hooks but their shape, the absolute size of the cirrus sac (smaller) and the triangular arrangement of the testes distinctly separate it out from these species. The typical shape of the ovary is another feature not shared by other forms.

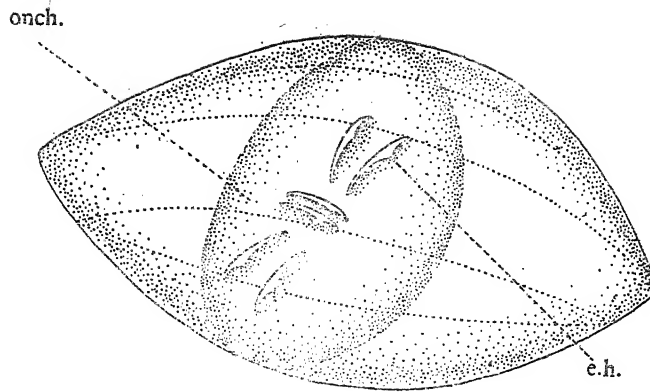


Fig. 3.

0.02 mm.

Egg of *Neeligerichis alternatus*

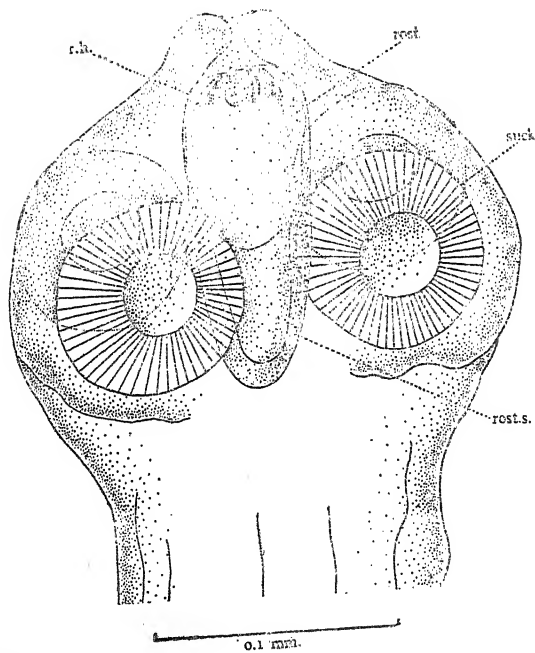


Fig. 4

Scolex of *H. ciconia* n. sp.

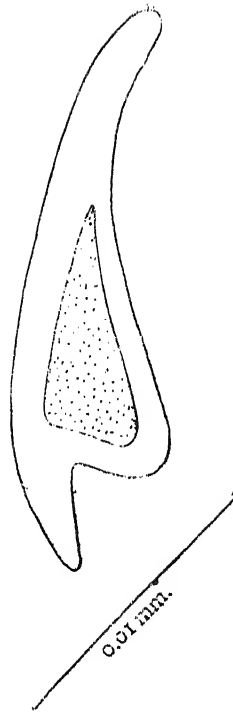


Fig. 5

Rostellar hook of *H. ciconia*.

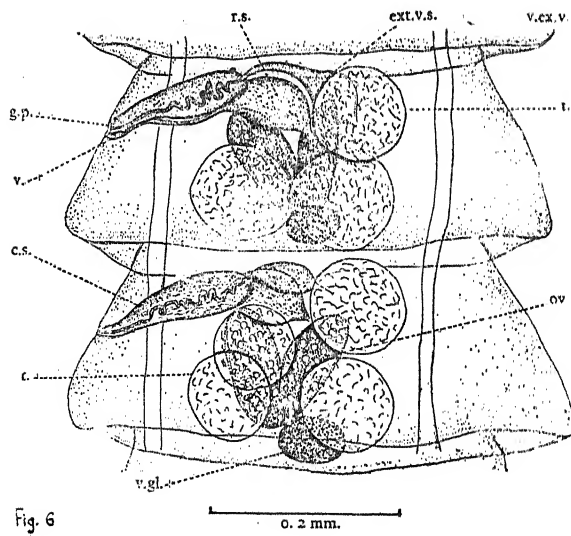


Fig. 6

Mature segment of *H. ciconia*

Hymenolepis graeca n. sp.

One complete and another incomplete worms were obtained from the intestine of a hill partridge *Alectoris graeca* Meisner in the neighbourhood of Tanakpur.

Length of the specimens 43 mm. and 69 mm., maximum width 0.56 mm. Scolex small, muscular 0.22 mm. \times 0.27 mm. Rostellum (0.12 mm. \times 0.05 mm.) armed with a single row of 8 hooks, 0.024 mm. long, blade being much smaller than the handle and curved, guard small and narrow but prominent. Rostellar pouch 0.2 mm. \times 0.09 mm. extending beyond the lower level of the suckers. Suckers globular, 0.11 mm. in diameter. Neck small 0.4 mm. long. All segments broader than long. Immature 0.1 mm. \times 0.2 mm. Mature and gravid ones 0.16 mm. \times 0.50 mm. and 0.24 mm. \times 0.54 mm. respectively. Last few segments almost squares 0.4 mm. \times 0.45 mm. Dorsal and ventral longitudinal excretory vessels 0.0056 mm. and 0.012 mm. in diameter respectively. Genital pores unilateral, at anterior $\frac{1}{4}$ of the lateral margin of the segments. Cirrus sac (0.10-0.11 mm. \times 0.045-0.05 mm.) globular crossing poral longitudinal excretory vessels and about $\frac{1}{4}$ across segment. Cirrus simple and unarmed. External vesicula seminalis large 0.07 mm. \times 0.059 mm. Testes (diameter 0.09 mm.) 3, two aporally one behind the other and the third poral adjacent to posterior aporal one. Ovary (0.12 mm. across) in posterior half of segment. Vitelline gland almost spherical 0.04 mm. in diameter. Receptaculum seminis large, maximum diameter 0.08 mm. Uterus saccular. Eggs numerous, diameter 0.09 mm., onchospheres 0.043 mm. and embryonic hooks 0.022 mm. long.

Amongst all the species of the genus *Hymenolepis* possessing 8 rostellar hooks the present form appears to resemble *H. lateralis* Mayhew, 1925; *H. incognita* Meggitt, 1927 and *H. filta* Meggitt, 1933. The location of all the testes poral to ovary, the relative size of the cirrus sac, smaller eggs and a large rostellum in the first; relative and absolute size of the cirrus sac, larger rostellar hooks and a very small strobila (5.7 mm. long) in the second and a smaller strobila with a small scolex possessing smaller rostellum bearing slightly bigger rostellar hooks in the third clearly separate these species from the present form. In addition, the shape of the rostellar hooks of all these species is also, in comparison, different.

Hymenolepis tanakpuria n. sp.

A very large number of worms were collected from the intestine of a hill partridge, *Alectoris graeca* Meisner in the neighbourhood of Tanakpur.

Length of the specimens 6-10 mm., maximum width 0.25 mm. Number of segments approximately 200. Margins of strobila smooth and strobilation faintly marked. Scolex (0.12-0.13 mm. \times 0.14-0.17 mm.) having a narrow apical region with suckers and rostellum and a broad basal region. Rostellum unarmed 0.04 mm. \times 0.028 mm. Suckers diameter 0.045-0.053 mm. Neck distinct 0.07-0.12 mm. \times 0.09 mm. Segments all broader than long. Immature 0.047-0.07 mm. \times 0.08-0.09 mm., mature ones 0.06-0.07 mm. \times 0.12-0.13 mm. and gravid ones 0.12-0.14 mm. \times 0.19-0.25 mm. Genital pores unilateral, dextral and near the middle of the lateral margin of the segments. Ventral longitudinal excretory vessel 0.006 mm. in diameter. Genital ducts situated dorsal to poral longitudinal excretory vessels. Cirrus sac (0.068-0.080 mm. \times 0.015-0.019 mm.) extends slightly diagonal towards the anterior border of the segment and beyond the middle of it. External vesicula seminalis diameter 0.025-0.031 mm. Internal one small. Testes (diameter 0.035-

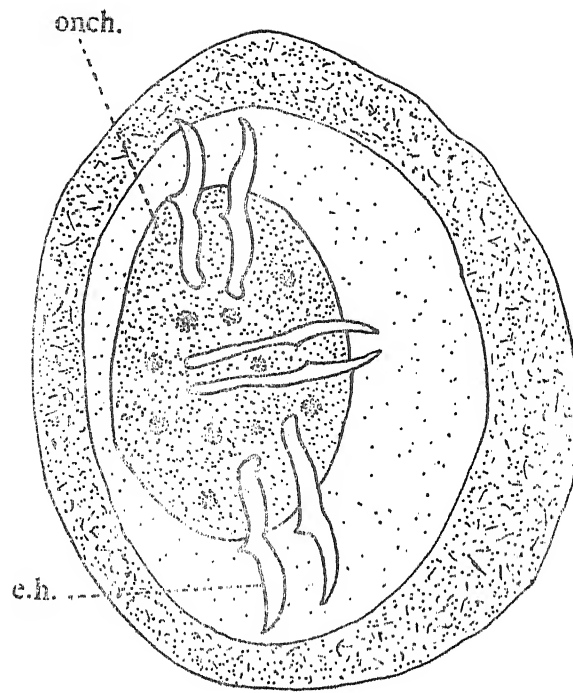
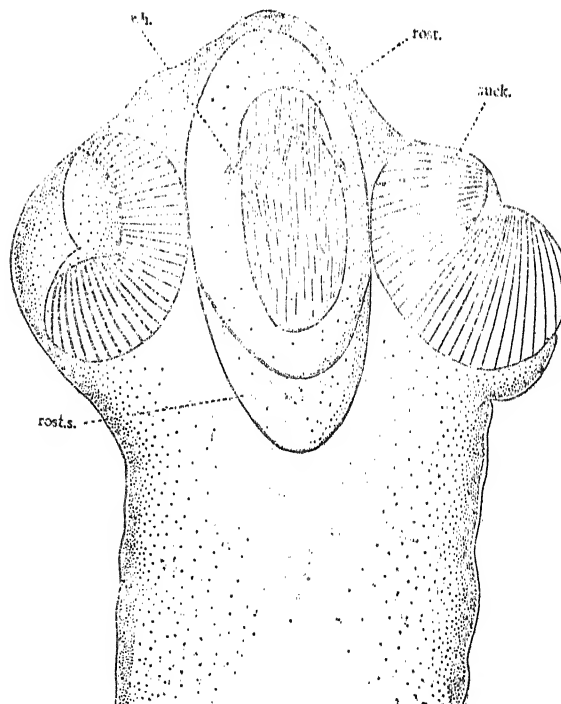
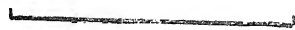
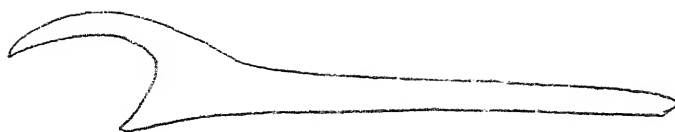


Fig. 7.

0.02 mm.

Egg of *H. cionina*





0.01 mm.

Fig. 9. Rostellar hook of *H. graeca*

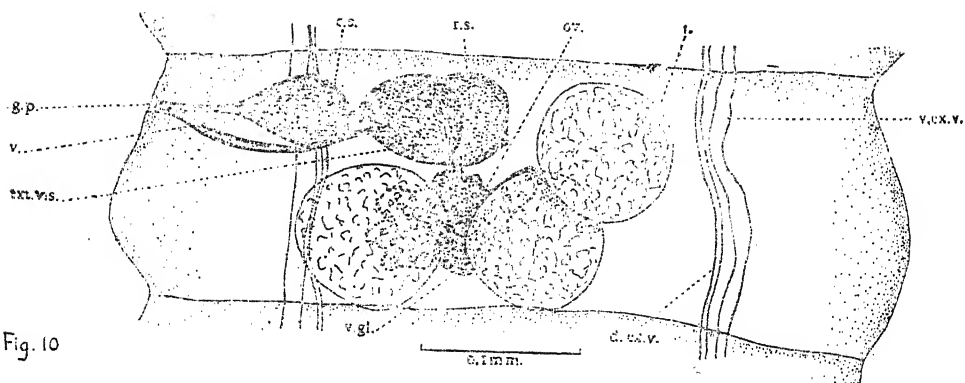


Fig. 10

Mature segment of *H. graeca*

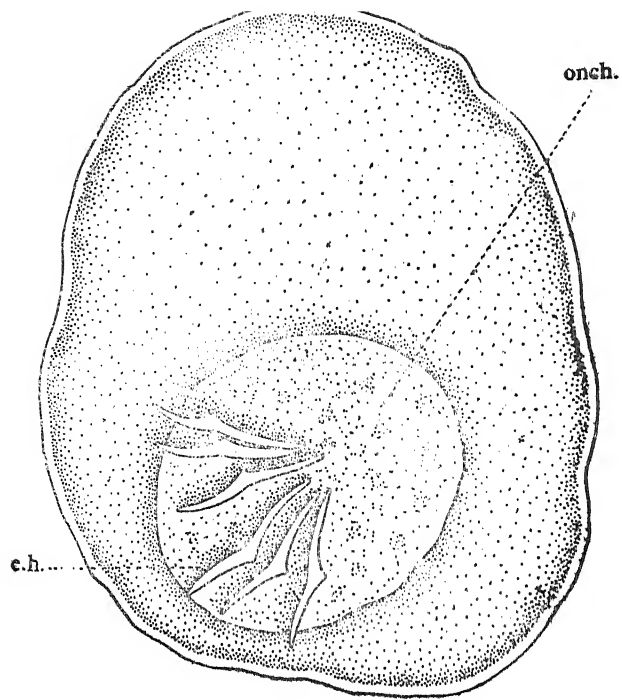


Fig. 11

0.05 mm.

Egg of *H. graeca*

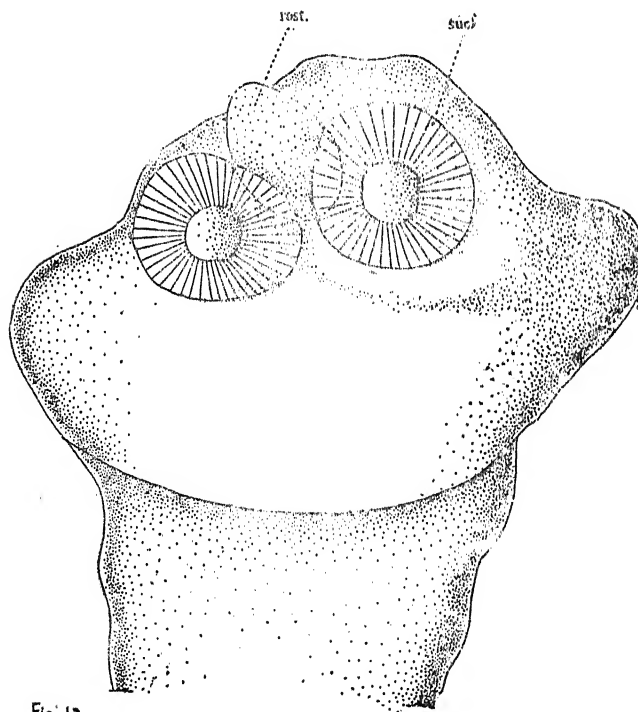


Fig. 12

0.05 mm.

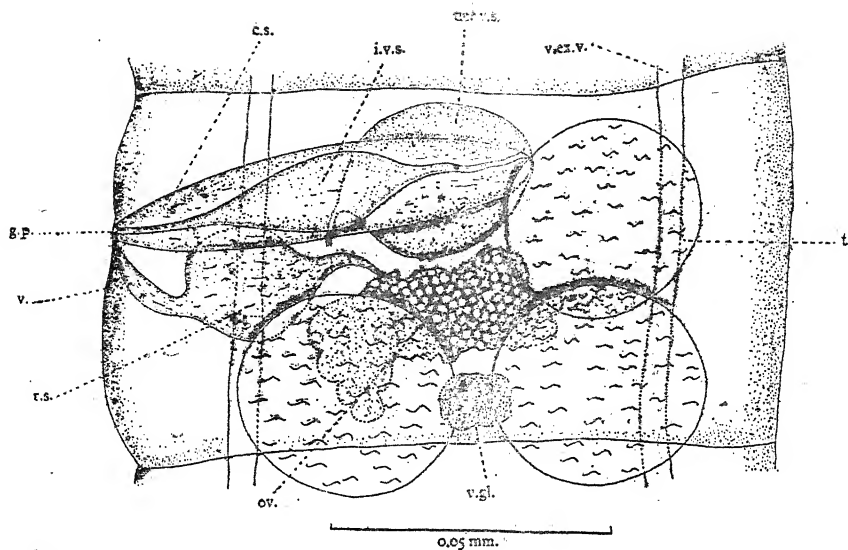


Fig. 13

Mature segment of *H. tanakpuria*

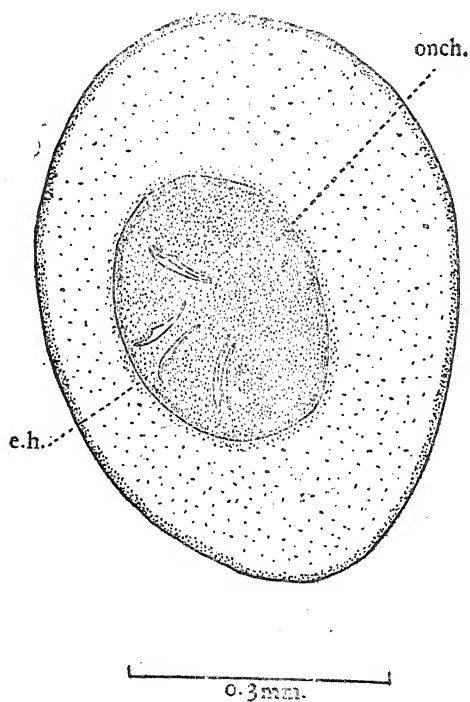


Fig. 14

Egg of *H. tanakpuria*

Abbreviations used

c. s., cirrus sac; d. ex. v., dorsal longitudinal excretory vessel; e. h., embryonic hook; ext. v. s., external vesicula seminalis; g. cl., genital cloaca; g. p., genital pore; i. v. s., internal vesicula seminalis; m., muscles; onch., onchosphere; ov., ovary; r. h., rostellar hook; rost., rostellum; rost. s., rostellar sac; r. s., receptaculum seminis; suck., sucker; t., testis; v., vagina; v. ex. v., ventral longitudinal excretory vessel; v. gl., vitelline gland.

0.040 mm.) 3, two aporal one behind the other and the third adjacent to the posterior aporal one. Ovary composed of cluster of small, irregular lobes near the centre of the segment, 0.04-0.06 mm. \times 0.02-0.028 mm. Vagina posterior to cirrus sac. Receptaculum seminis almost spherical, diameter 0.020-0.028 mm. Uterus saccular occupying whole of gravid segments. Eggs numerous, slightly oval 0.05-0.06 mm. \times 0.067 mm. Onchospheres 0.024 mm. \times 0.03-0.032 mm. Embryonic hooks 0.008 mm. long.

The present form comprising extremely small specimens having an unarmed scolex is easily distinguished from all the hitherto known species excepting *H. alpestris* Baer, 1931, *H. globosa* Baer, 1931 and *H. rustica* Meggitt, 1926. However, it can be separated from *H. alpestris* and *H. globosa* by the possession of a smaller scolex with smaller suckers. Further, *H. alpestris* has a larger rostellum with smaller number of eggs (10) in the uterus and *H. globosa* is extremely small (1.3-2.0 mm. long) and, therefore, both of them stand apart from the present form. From *H. rustica* the present form is separated in possessing a comparatively smaller cirrus sac which extends beyond the centre of the segment.

The author wishes to express his grateful appreciation to Professor M. B. Lal under whose guidance the present investigations were carried out.

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THE MORPHOLOGY OF THE MALE REPRODUCTIVE ORGANS OF

Pantala Flavescens FABRICIUS (Libellulidae: Odonata)

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INTRODUCTION

The earliest reference of the work on the morphology of the reproductive organs of dragonflies dates as far back as 1832, the year in which Rathke published a paper viz. "De Libellarum partibus genitalibus." In 1896 Martha Freeman Goddard described the secondary copulatory apparatus in *Diplax rubicundula*, *D. vicina*, *Celithemis elisa*, *Libellula pulchella*, *L. quadrupla*, *L. exusta* and *Plathemis trimaculata*. The development of the appendages of the second abdominal segment of male dragonflies has been worked out by Thompson (1908). E. W. Roberts published a very interesting paper in 1912 according to which the intromittent organ of the Odonata was homologous with a pair of legs fused to form a median structure. Kennedy (1922) gives a good account of the external and internal anatomy of the penis especially of the genus *Libellula*. The complete external morphology of the imago of the dragonfly, *Onychogomphus ardens* Needham has been described by Chao (1953) and that of the dragonfly larva, *Anax junius* Drury by Snodgrass (1954). The morphology and histology of the internal reproductive organs of dragonflies has been described only by Marshall (1914) in the genus, *Libellula quadrimaculata* Linn. and Tillyard (1917) in *Aeschna*. From the above it is evident that very little work has been done on the reproductive system of dragonflies and practically no work has been done on the Indian dragonflies.

MATERIAL AND METHOD

The adults of *Pantala flavescens* Fabricius were captured with a large net mostly during the months of August and September. These were dissected in 0.75% sodium chloride solution and the reproductive organs fixed for 18-20 hours in a mixture of Picro-Formol-Acetic Acid in the ratio 3:1:0.2. Serial sections of the fixed material, cut 6-8 micra thick on a Cambridge Rocking Microtome, were first stained either with Delafield's Haematoxylin or Heidenhein's Haematoxylin and then counter stained with Eosin. For preparing whole mounts of the external genitalia, the portions bearing the organs were cut and treated in 10% KOH solution for 4-6 hours in a bath at 60°C temperature. These were washed thoroughly with water, dehydrated and mounted unstained.

ACKNOWLEDGMENTS

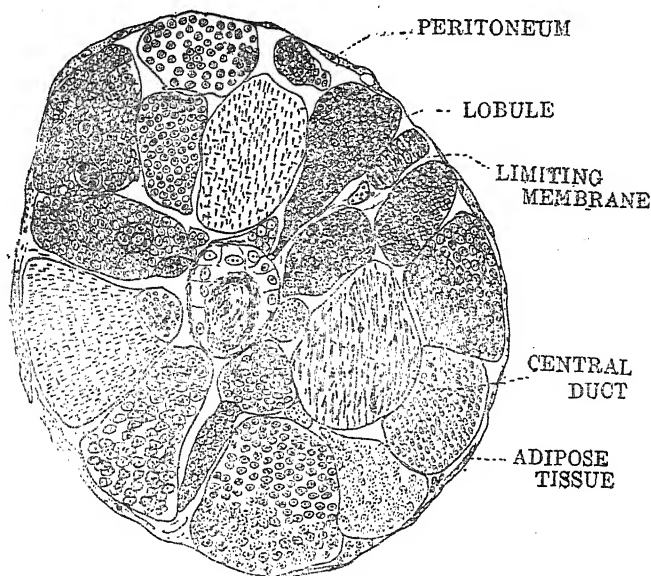
The authors are greatly indebted to Prof. M. D. L. Srivastava of the Zoology Department, Allahabad University for providing the necessary facilities for the completion of the work. They are also thankful to Mr. D. E. Kimmins of the British Museum, London for getting the specimens identified.



- Fig. 1. Microphotograph of the dissection showing male internal reproductive organs.
 Fig. 2. T. S. of Testis.
 Fig. 3. Photomicrograph of vas deferens. (45·5x0 X 10x E).
 Fig. 4. Photomicrograph of section of sperm sac. (10x 0 X 10x E).
 Fig. 5. Photomicrograph of section of sperm sac showing ejaculatory duct. (10x 0X10x E)
 Fig. 6. Photomicrograph of a highly magnified portion of the wall of sperm sac. (45·5x0 X 10x E)

GROSS MORPHOLOGY OF THE INTERNAL GENITAL ORGANS

The internal genital organs in male *Pantala flavescens* Fabr. consist of a pair of testes, a pair of ducts, the vasa deferentia, and a sperm sac. Accessory glands are totally absent.



Text Fig. 1. Figure showing transverse section of testes.
(Camera lucida sketch of the cross section of the testis).

The testes:—There are two testes each lying on either side of the alimentary canal. Each testis is an elongated, translucent (in living state), cylindrical, tubular and multifollicular organ consisting of a very large number of lobules. It is situated nearly in whole of the sixth and seventh abdominal segments (TS) (Plate I, fig. 1). The length and position of the testis varies greatly in different individuals according to the development of the organ but remains within limits described above. Testes are held in position by tracheae, fat-bodies and the anterior filaments.

The Vas Deferens:—Posteriorly each testis is attached terminally to a duct, the vas deferens, running latero-ventrally to the alimentary canal and extending from the beginning of the eighth abdominal segment upto the middle of the ninth abdominal segment (VD) (Plate I, fig. 1). Its average length is 0.5307 millimetres. The vas deferens gradually swells up towards the posterior side, and in the middle of the ninth abdominal segment forms a small loop before joining its fellow from the opposite side. The loop is very weakly defined or may be absent altogether. The vas deferens is, therefore, divisible morphologically into two portions; (i) Anterior thin portion lying immediately after the testis and (ii) comparatively thick and enlarged posterior portion.

The Sperm Sac:—The short common duct formed by the union of the two vasa deferentia, immediately swells up on the dorsal side to form a more or less spherical sac-like structure, the sperm sac (SS) (Plate I, fig. 1), lying beneath the hind-gut and measuring 0.459 mm. in antero-posterior direction and 0.51 mm. across. The ventral side of this sperm sac rests in a depression formed by the

invagination of the wall of male gonopore. The two vasa deferentia open straight into the sperm sac on the anterior side.

The Ejaculatory Duct:—The ventral portion of the sperm sac resting in the chitinous pit is modified into an ejaculatory duct (ED) (Plate I, fig. 5) which is microscopic and has the form of a deep cup-like pit. The ejaculatory duct opens out to the exterior through a longitudinally elongated slit-like male gonopore.

HISTOLOGY OF THE INTERNAL GENITAL ORGANS

The Testes:—Each testis is surrounded by adipose tissue and peritoneum (Plate I, fig. 2). The adipose tissue does not form a complete layer round the testis. It consists of loose groups of fat-bodies attached irregularly. This tissue consists of large fat cells with big granular and round nuclei enclosed within highly vacuolated cytoplasm. Many dark staining granules are seen scattered here and there in the fat cells. It is richly supplied with tracheoles. Peritoneum consists of flat cubical cells provided with large nuclei. A few of the nuclei contain deeply staining granules also. Peritoneum is quite distinct in this animal and at some places it becomes quite thick while at other places it is extremely thin. Peritoneal layer forms a complete covering round the testis.

A very large number of lobules is contained inside the wall of the testis (L) (Plate I, fig. 2). The lobules are connected to a common central duct running in the middle of the testis, throughout its length. Lobules filled with spermatids and spermatozoa are found on all sides both at the periphery and near the central duct thereby showing an irregular arrangement. The wall of the central duct is composed of an inner epithelial layer surrounded externally by a muscle layer. The epithelial layer consists of a single row of closely packed cubical cells, each with a large nucleus. The nucleus contains a prominent nucleolus and occupies nearly the whole of the space at the base of the cell. Muscle layer is formed by a single row of circular muscle fibres arranged round the duct. A basement membrane between the epithelial layer and muscle layer is not distinct.

Structure of the Lobules:—A lobule is almost a spherical and solid mass of germ cells bounded externally by a very thin limiting membrane. Each lobule contains a large number of germ cells (G) (Plate I, fig. 2) in a particular stage of spermatogenesis. Different lobules contain germ cells in different stages of spermatogenesis. A section of the testis from any place shows the lobules containing spermatogonia, spermatocytes, spermatids and even mature spermatozoa. The germ cells in a single lobule are all alike and there is no indication of a germinal epithelium. Moreover the lobules are solid having no space in the centre. The lobules open into the central duct by very minute ductules formed seemingly by the prolongation of the lobule itself as they have the same histological structure as the lobules.

The Vas Deferens:—Histologically also the vas deferens is divided up into two regions which correspond to the morphological divisions:—

(i) *Anterior thin region*:—The epithelial layer of the wall of the vas deferens in the anterior thin region consists of a single tier of triangular, elongated and cubical cells with big nuclei which occupy nearly the whole of the basal space of the cell. Each nucleus contains a prominent nucleolus while 4-5 deeply staining small granules are also seen attached to the nuclear membrane. All the cells of the epithelial layer are of one type. Next to the epithelium on the outer side is a prominent and thick layer of adipose tissue which forms more or less a complete surrounding layer. The adipose tissue is mainly composed of typical fat cells and is richly supplied with finer

branches of tracheoles. In between the adipose tissue layer and the epithelium is present a thin cellular layer, the basement membrane on which the basal ends of the epithelial cells rest. In the centre there is a spaceous cavity which is not lined by chitinous intima but contains mature and immature spermatozoa. The diameter of the vas deferens in this region is 0.0504 mm. which is exclusive of the layer of fat-bodies whose thickness is variable.

(ii) *Posterior region* :—Posterior to the region described above, the vas deferens gradually swells up and attains a maximum diameter of 0.1224 mms. Then again near the region of sperm sac the diameter of the duct diminishes to 0.0792 millimetres. All these measurements exclude the thickness of the adipose tissue layer. Following layers are present in the wall of the posterior region of vas deferens from within outwards (Plate I, fig. 3) :—

(A) Epithelial layer, (B) Basement memberane, (C) Circular muscle layer and (D) Adipose tissue layer.

The epithelium (EP) consists of a single row of laterally compressed and elongated columnar cells containing rounded nuclei at the bases. The nucleus shows a distinct nucleolus. The muscle layer (CM) is very thin, composed of circular muscle fibres which, however, form a strong muscular coat in the region of the vas deferens near the sperm sac. There is a very thin cellular basement membrane interposed between the epithelial layer and the muscular layer. Adipose tissue layer (AT) is quite thick and well developed in this region and consists of typical fat cells. In the centre, the organ contains a lumen which varies in size. When epithelial cells are well developed and greatly elongated, the cavity is reduced to a small space in the centre and may or may not contain sperms. But, when the cavity is greatly developed, the epithelial cells are small and form a comparatively thin lining round the cavity. There is no chitinous intima in this region of the vas deferens.

The Sperm Sac. :—The orientation of the tissue in the sperm sac is as follows (Plate I, figs. 4 & 6)—(i) Epithelium, (ii) Basement membrane, (iii) Musculature and (iv) Adipose tissue layer.

The epithelium of the sperm sac is greatly developed and is thrown internally into many irregular folds (*f*) which are distinctly visible when the sperm sac is not distended. The epithelial layer consists of columnar gland cells in which the nuclei are present near the free ends (Plate I, fig. 6). The basal ends of the epithelial cells rest on a very thin cellular basement membrane which is visible with difficulty. The musculature (MS) is very well developed in this organ and consists of striated circular muscle fibres arranged in several rows. The muscular coat forms a thick covering round the wall of the sperm sac. Adipose tissue layer consists of typical fat cells and is also very well developed forming a thick layer round the organ. In the centre, the sperm sac contains a spaceous cavity which stores spermatozoa, and is not lined by chitinous intima.

The Ejaculatory Duct :—The ejaculatory duct (ED) (Plate I, fig. 5) in *Pantala flavescens* Fabr. is comparatively smaller than that found in other species and shows histologically the same structure as the sperm sac except that its epithelium is lined internally by a very thin chitinous intima which is broken at places.

EXTERNAL GENITAL ORGANS

Male external genitalia of *Pantala flavescens* Fabr. consists of a pair of supra-anal appendages, an unpaired infra-anal appendage, a pair of small appendages on the ninth abdominal sternum and a secondary copulatory apparatus on the ventral

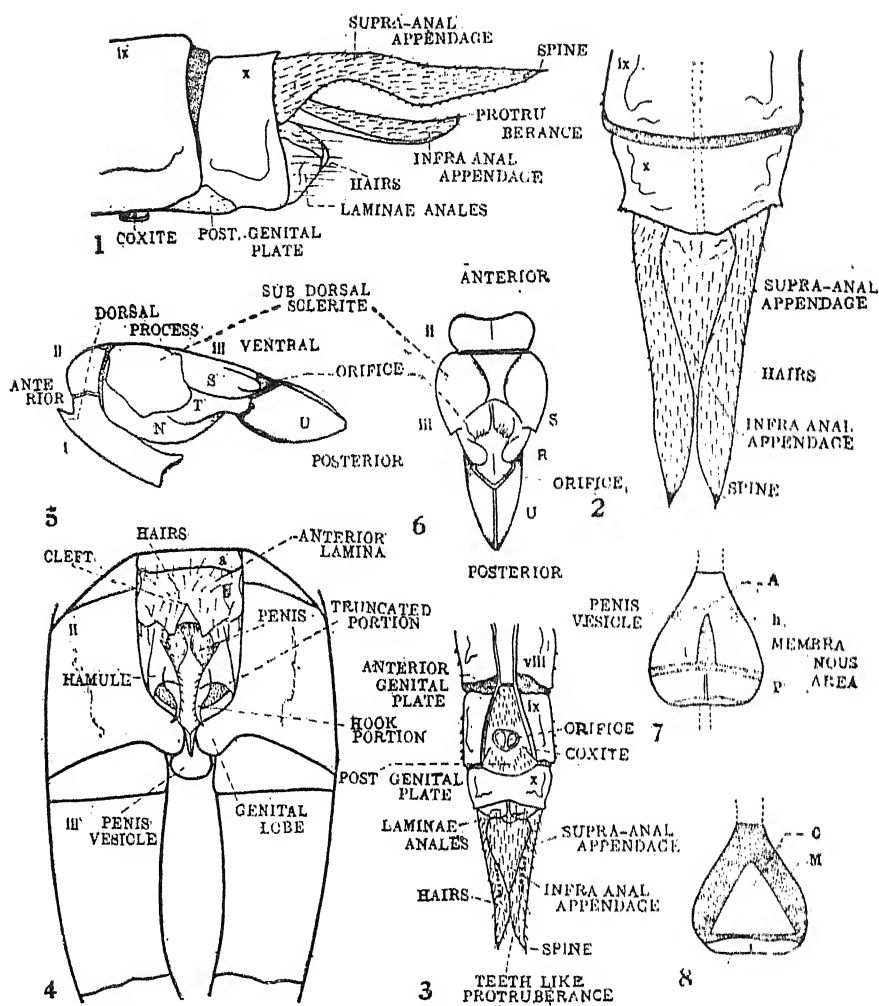


PLATE II

(All diagrams have been sketched using 1.5 x Objective lens and 10 x Eye-piece)

- Fig. 1. Lateral view of abdominal segments IX and X.
- Fig. 2. Dorsal view of abdominal segments IX and X.
- Fig. 3. Ventral view of abdominal segments VIII, IX and X.
- Fig. 4. Ventral view of secondary copulatory apparatus.
- Fig. 5. Lateral view of penis. (2.0 x 0 X 10 x E)
- Fig. 6. Dorsal view of penis. (2.0 x 0 X 10x E)
- Fig. 7. Ventral view of Penis vesicle
- Fig. 8. Dorsal view of Penis vesicle

side of the second and a part of the third abdominal segments. The male genital opening is situated in the mid-ventral line on the ninth abdominal segment.

The Supra-anal Appendages:—There is a pair of long, cylindrical, forceps-like supra-anal appendages coming out dorso-laterally from the terminal end of tenth abdominal segment and bearing a minute black spine at the apex (Plate II, fig. 2). Each supra-anal appendage is more or less flat at the base but is tubular distally and measures 4.038 mm. in length. The proximal one third portion of each appendage is pale yellow in colour, while the rest of the appendage is black. Numerous bristle-like hairs, situated on small protruberances, are found all over the surface of the supra-anal appendage except the 0.068 mm. long spine area. Each supra-anal appendage bears on the mid-ventral line a large number of small black tubercle-like protruberances which are arranged in a row.

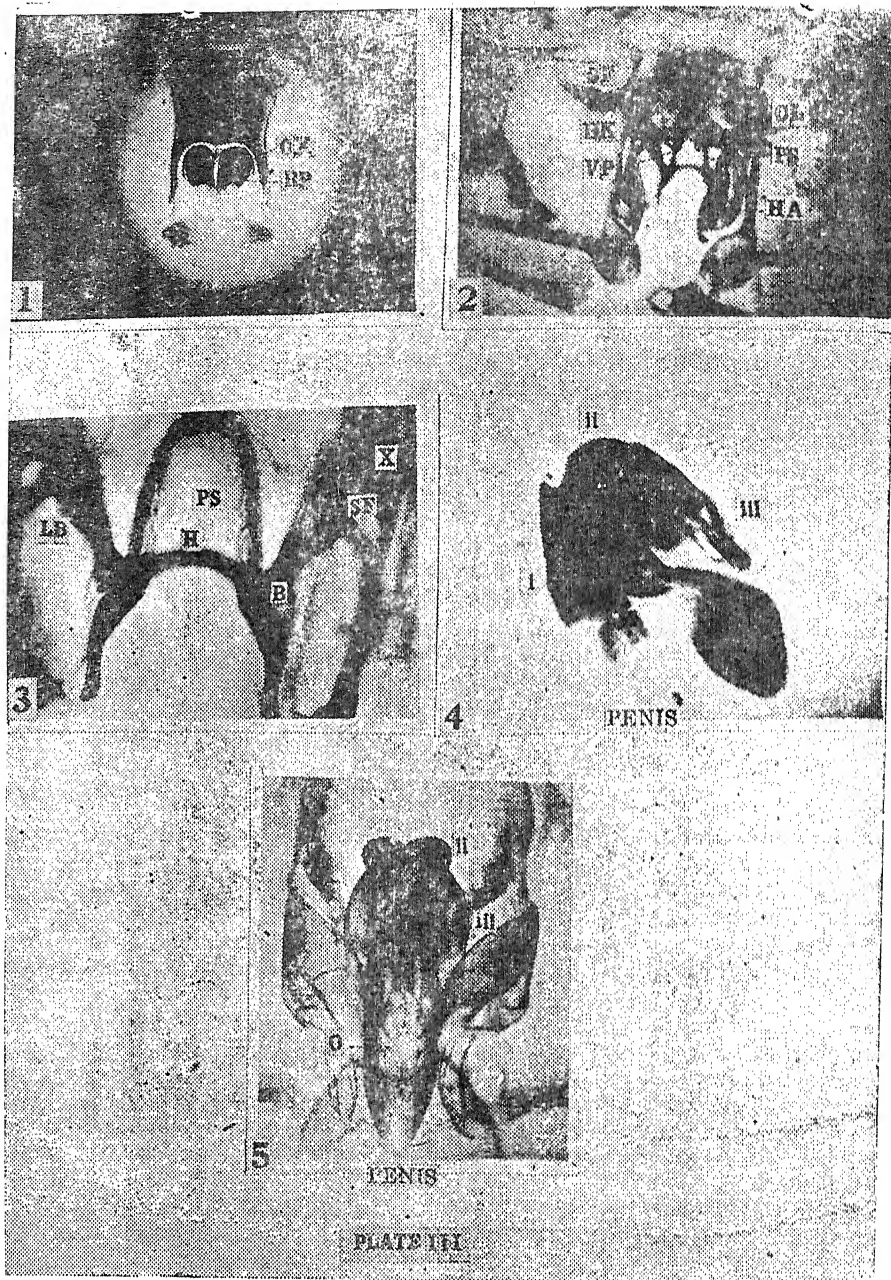
The Infra-anal Appendage:—Situated ventral to the supra-anal appendages and dorsal to the anus is a median, unpaired, spoon-shaped chitinous piece, the infra-anal appendage, a little more than half of the length of supra-anal appendage (Plate II, fig. 1). The infra-anal appendage is narrow proximally, broad in the middle and tapering distally with a depressed dorsal side. It is characteristically present only in the males. On the dorsal side of the tip it bears two symmetrical black protruberances on either sides of a faintly indicated middle line. This indicates the paired nature of the infra-anal appendage, the symmetrical pieces being fused together mid-ventrally. Large hairs are uniformly present on the ventral side of infra-anal appendage, but the dorsal surface is devoid of hairs, except the apical region where few small hairs are found. The infra-anal appendage bears a depression proximally on the ventral side into which the median lamina supra-analis fits. The infra-anal appendage is pale yellow with margins and apical region coloured black.

The Male Gonopore:—Male genital opening is situated in the middle of the ventral side of the ninth abdominal segment (Plate II, fig. 3 and Plate III, fig. 1). It is a longitudinally elliptical slit 0.289 mm. long and is guarded by a pair of small appendages, the coxites (CX). The coxites are thick somewhat elongated and oval structures measuring 0.612 mm. in length, the tapering end being directed posteriorly. Posterior to the genital opening is a chitinous post-genital plate, which is rounded apically and seems to develop as a result of secondary sclerotization of the intersegmental area behind the genital pore. This plate is adorned by many hairs. The portion of the ninth abdominal sternum lying anterior to the genital opening is modified to form an elongated anterior genital plate, 0.68 mm. long. It is beset with numerous minute tubercles and a few small hairs. The chitinous wall of the male genital pore is drawn up dorsally to form a large triangular shallow cup-shaped structure (RP) (Plate III, fig. 1) measuring 0.51 mm. in length and 0.476 mm. in width. It bears hairs on the ventral side and lodges the ejaculatory duct.

THE SECONDARY COPULATORY APPARATUS

The secondary copulatory apparatus (Plate II, fig. 4) in male *P. flavescens* Fabr. consists of anterior and posterior laminae, genital fossa, supporting framework, hamules, penis sheath, penis vesicle, penis and the genital lobes which are typical of the family Libellulidae.

The Genital Lobes:—These are a pair of comparatively short (0.552 mm. long) and apically rounded processes situated posterior to the hamules. The genital lobes are projections from the postero-lateral region of the tergum of the second abdominal segment which normally lie folded over the penis vesicle at right angles to the plane



- Fig. 1. Photomicrograph of the 9th sternum showing coxites, gonopore and genital plates etc. (1 x 0 X 10 x E).
- Fig. 2. Photomicrograph of secondary copulatory apparatus. Penis removed (ventral view) (1x 0 X 6x E).
- Fig. 3. Photomicrograph (close-up) showing penis sheath, supporting framework, and the bases of hamules. (10x 0 X 6x E).
- Fig. 4. Photomicrograph of penis (permanent chitin preparation). (Lateral view). (1x 0 X 10x E).
- Fig. 5. Photomicrograph of penis showing the opening (Dorsal view). (1x 0 X 10x E).

of abdomen. These bear large hairs near the margin and on the ventral surface and assist in the erection of penis.

The Genital Fossa :—On the ventral side of the second abdominal segment is formed a membranous groove or the genital fossa. All other secondary copulatory organs formed by the modification of second abdominal sternite and the penis lie in this fossa. Anteriorly the fossa is bounded by anterior lamina, posteriorly by posterior lamina and laterally the fossa is strengthened by the chitinous framework. The sheath of the penis (PS) (Plate III, fig. 2) occupies an area which is greater than the posterior half of the genital fossa. The genital fossa does not open posteriorly into the penis vesicle as stated by Tillyard (1917).

The Anterior Lamina :—It is a large chitinous sclerite lying ventrally in the anterior half of the second abdominal segment. It is distinctly differentiated into two parts, a small anterior (a) narrow, almost flat and transversely elongated plate and a large posterior (b) convex hood-like portion. The upper (ventral) surface of the latter is beset with numerous large hairs. The posterior portion of the anterior lamina is produced laterally and dorsally to form a basket whose base is formed by the membrane of the genital fossa. The posterior margin of the anterior lamina is regular, deeply cleft in the meso-ventral line (CL) (Plate II, fig. 4 and Plate III, fig. 2), and is drawn out laterally into two small hook-like structures (HK). On the two sides of the median cleft of the anterior lamina are found two prominent tongue-shaped processes, the ventral processes (VP) of the anterior lamina, directed ventro-posteriorly. These overlap the anterior portion of the reflexed penis. The anterior lamina extends over the anterior one third part of the penis sheath containing the penis.

The Posterior Lamina :—It is a distinct but weakly chitinized piece lying at the posterior end of the genital fossa in the second abdominal segment. It is narrow in the middle but expanded laterally. Usually it lies concealed under the hamules and the anterior region of the penis vesicle.

The Supporting Framework :—The hamules are borne by a lateral chitinous framework (SF) (Plate III, fig. 3) roughly U-shaped in structure which also gives support to the genital fossa, penis sheath and anterior lamina. This framework consists of chitinous rods which are formed by local chitinization of the membrane of that region (Goddard, 1896). The anterior ends of the 0.586 mm. long lateral bars (LB) of U-shaped framework are attached to the under and inner sides of the posterior portion of anterior lamina near its antero-lateral basal end. The horizontal rod (H) of the U-shaped framework is curved in the middle to support the penis sheath from below. Two very small chitinous processes (X), one on each side, are given out from the lateral arm of 'U', a little anterior to the horizontal depressed bar of the framework. These processes project ventrally and form the anterior point of attachment for the hamules.

The Sheath of the Penis :—It is a chitinous scoop-like structure roughly triangular in shape, measuring 1.02 mm. in length (Plate III, fig. 3). It has a broad shallow longitudinal cavity, which is open on the ventral side and lodges the penis. The apex of the triangle is rounded and points anteriorly, while the bases are directed posteriorly and are joined together by a thin chitinous line. The lateral arms (B) of the triangle are notched near the middle, and posterior to the horizontal bar of U-shaped framework, seem to bulge apart from each other by turning laterally and posteriorly. The anterior pale-brownish region of the penis sheath bears hairs on the dorsal and lateral sides. Hairs are also present on the apex of the penis sheath. The sheath of the penis occupies more or less a central position on the ventral side of the second

abdominal segment. The posterior basal ends of the triangular penis sheath form the posterior point of attachment for the hamules.

The Hamules :—A single pair of stout appendages (HA) which are easily seen even with the naked eyes, are found laterally in the genital fossa lying anterior to the genital lobes and posterior to the anterior lamina (Plate II, fig. 4 and Plate III, fig. 2). Each hamule is a thick, laterally compressed and somewhat elongated organ with a concave proximo-basal end, which is strongly chitinized laterodorsally. Distally the hamule is divided into two parts: an anterior large, dark-brown strongly chitinized spur-like portion bearing a short black hook at the tip, and a posterior short, brown, truncated triangular portion. The face of the truncated portion is turned towards the concave side of the spur-like portion, whose outer margin bears numerous long hairs. The inner side of the truncated lobe also bears small blunt hairs. The anterior inner lobe of the hamule is larger in length than the truncated part. The spur-like lobe is directed laterally and ventrally while the truncated lobe points laterally and posteriorly. The basal portion of the hamule is weakly chitinized on the mesal face. Each hamule is attached anteriorly to the framework and posteriorly to the posterior basal ends of the penis sheath. The hamules maintain proper positions during copulation.

The Penis Vesicle :—Penis vesicle is a large, hemispherical flask-like body measuring 1.19 mm. in length (Plate II, figs. 4, 7 and 8). It has a flat bottom which is drawn into the cavity near the middle. The dorsal plane surface of the penis vesicle is membranous and is attached for a considerable distance to the underlying part of the abdomen and bears meso-posteriorly a triangular chitinized sclerite (C) (Plate II, fig. 8). The ventral surface bears three distinct strongly sclerotized sclerites, one postero-median (P) and the other two antero-lateral (A) in position (Plate II, fig. 7). The postero-median sclerite is a band-like and transversely elongated chitinous piece covering the posterior basal portion of the vesicle. All these sclerites are separated from each other by membranous interspaces which allow the distention of the penis vesicle. On the ventro-lateral side of the neck or the anterior tapering region of the penis vesicle is a raised area which bears cluster of hairs (h) situated on protruberances. The cavity of the penis vesicle is filled up with a fluid and mature spermatozoa, which are transferred to the bursa copulatrix of the female during copulation. The penis vesicle does not open into the genital fossa anteriorly, nor does it open into the haemocoel.

The Penis :—Penis in *Pantala flavescens* Fabr. is a very complicated median segmented rod-like organ attached terminally to the neck of the penis vesicle and is located ventrally in the second abdominal segment (Plate II, fig. 4). Penis is three jointed and its distal segment is flexed over the proximal (Plate II, fig. 5 and Plate III, fig. 4). The first segment is dorsal, second one is terminally antero-ventral and the third segment is ventral in position.

The first or the proximal segment is a simple stout rod slightly curved ventrally and measuring about 0.816 mm in length (Plate II, fig. 5). It is strongly chitinized dorsally and laterally but on the ventral side over which lies the reflexed distal segment, it is very weakly sclerotized. It bears subapically on the dorso-distal side, a prominent triangular strongly sclerotized process, the dorsal process. The cavity of this segment is continuous with that of the penis vesicle. This segment of the penis lies in the groove of the penis sheath and is directed anteriorly.

The second or the middle segment of the penis is smallest of the three measuring 0.391 mm. in length and lies perpendicularly antero-ventral to the distal end of the first segment. It is strongly chitinized laterally and sub-dorsally but meso-dorsally it

is membranous. It is roughly triangular in shape with rounded anterior dorsal border. On the antero-ventral side it has a narrow surface which is membranous.

The third or the distal segment is largest measuring about 1.7 mm. in length (Plate II, fig. 5 and Plate III, fig. 4). It has a very complicated structure consisting of several sclerites. Proximally, there is a pair of strongly sclerotized sub-dorsal sclerites, one on each side of the median line (Plate II, fig. 6). These are broad anteriorly at the base and narrow posteriorly. Postero-dorsal to these is another pair of sclerites whose distal margin is very irregularly indented (S). Arising laterally from the sub-dorsal sides is a pair of elongated sclerites which are produced distally into rounded lobes (R). The rounded lobe-like structures tend to meet each other mid-dorsally. Further postero-distally, two broad triangular and laterally compressed chitinous pieces (U) are placed together in such a manner that they form a long, narrow mid-dorsal groove which continues upto the tip of the penis. At the proximal end and on the latero-ventral side are found two chitinous plates (N), one on each side. Laterally there is a curved strongly chitinized sclerite (T) situated at about the middle of the segment. The ventral wall of the penis consists of a weakly sclerotized membrane. Near the middle on the dorsal surface is an orifice (O) (Plate III, fig. 5) which is guarded antero-dorsally by a pair of sclerites whose free margin is irregularly indented, and latero-dorsally by two lobes. This opening continues posteriorly through a groove enclosed by two laterally compressed and elongated chitinous plates upto the tip of the penis.

DISCUSSION

As described earlier, the dragonfly testis consists of a single long cylindrical organ with a wrinkled appearance. It is attached terminally to the vas deferens. Apparently the testis appears to be unifollicular in nature, but a study of the histological details reveals that the structure is rather otherwise. The lobules (or cysts of Marshall, 1914) containing developing germ cells (i.e. spermatogonia, spermatocytes, spermatids and spermatozoa) are arranged irregularly around the central duct. The different zones, i.e. those of germarium, maturation and transformation, characteristic of a typical testicular follicle are absent in the present case. Moreover each of the lobules of the dragonfly testis communicates with the central duct through a very minute ductule. Evidently enough in *P. flavescens* Fabricius the testis is not unifollicular in nature but is multi-follicular or multi-lobular*.

George (1928) mentioned the central duct running upto the middle region of the testis in *Agrion*. In the present case, on the other hand, it is found running through the entire length of the testis. Tillyard (1917) reported in *Aeschna* that before joining the sperm sac, each vas deferens formed loops. In *P. flavescens* Fabr. the vasa deferentia open straight into the sperm sac on the anterior side forming no loop whatsoever. This is probably due to the fact that in *Aeschna*, the sperm sac is comparatively less developed and the spermatozoa are stored in the vas deferens also, thereby increasing its length and forming loop. On the other hand no such necessity arises in *P. flavescens* Fabr. where the sperm sac is large and well developed and the vasa deferentia open straight into it without forming loops.

Marshall (1914) and Tillyard (1917) report that the sperms are present in bundles forming spermatophores inside the sperm sac. In the species under investigation no spermatophores have been found in the sperm sac. Special careful efforts made to find out the spermatophores in the vas deferens proved futile, a fact contrary to the finding

*According to Imms "Text-book of Entomology" 1951 Edition, lobules and follicles are synonymous.
Page 180.

of Tillyard (1917). Absence of the accessory glands is easily explained by the fact that the wall of the sperm sac is glandular.

The supra-anal appendages and the infra-anal appendage are copulatory organs used by the male in clasping the body of the female during mating. For this reason they are well-developed. The middle line and the pair of black protruberances situated at the dorso-distal tip of the infra-anal appendage of *P. flavescens* Fabr. indicate the originally paired nature of the appendage. This is further supported by the fact that the infra-anal appendages are paired in Zygoptera, a group primitive than the Anisoptera, to which *Pantala* belongs. The row of black teeth-like protruberances observed on the outer ventro-lateral side of the supra-anal appendages, point to the possibility that they are used in grasping the female more perfectly by providing a rougher surface. These teeth-like protruberances have not been reported by any worker so far except Snodgrass (1954), who has shown them only in the diagram, and is silent about these in the discussion. Since they have been observed occurring constantly only in the males, they seem to represent secondary sexual characters.

Owing to the transference of the copulatory function from the posterior region to the anterior region of abdomen, the second abdominal segment has become greatly modified. It is characterised by the presence of a very complicated set of organs forming the secondary copulatory apparatus, which is a distinctive possession of the male dragonfly only, having no parallel in the whole of the animal kingdom. Prior to copulation the sperms are transferred to the penis vesicle present ventrally in the second abdominal segment by flexing the abdomen downwards and forwards. For this reason the functional penis is situated in the second abdominal segment and not in the ninth abdominal segment. Snodgrass (1935)* has termed the intromittent organ in Odonata as "secondary penis", for, he believes that a reduced, but true penis exists in the ninth abdominal segment. George (1928) also holds a similar view. Apparently there seems to be no penis in the ninth abdominal segment. A close study, however, reveals a cup-like chitinous organ lodging the ejaculatory duct in its cavity, located into the ninth segment. Probably George and Snodgrass refer to this structure when they report the existence of a true penis, but neither of them has given its detailed description. Structurally this organ appears to be formed by the invagination of the ventral chitinous body wall through the male gonopore. It is inevitable and quite different from the typical penile form, but its situation and association with the ejaculatory duct lead us to conclude that it is the rudimentary true penis, that has assumed a peculiar and very different form as a result of degeneration. In the present description, however, the term penis has been used for the intromittent organ of *P. flavescens* Fabr. following the terminology of a majority of workers of Odonatology viz. Thompson (1908), Tillyard (1917) and Chao (1953) etc. The orifice of the penis in *P. flavescens* Fabr. is situated on the dorsal side of the third segment of the penis near its distal end, and not on the convex dorsal side of the second segment as described by Tillyard (1917) in *Aeschna*.

SUMMARY

1. The testes are a pair of cylindrical multifollicular organs, each extending on either side of the alimentary canal in whole of the sixth and seventh abdominal segments.
2. The testes are held anteriorly by a few filaments also besides being supported by tracheae, fat-bodies and nerves.

* "Principles of Insect Morphology" 1935 Edition.

3. Histologically the testis is surrounded by adipose tissue and a thin peritoneum enclosing a large number of solid and spherical lobules.
4. A lobule is surrounded externally by a thin limiting membrane only, there being no germinal epithelium in the adult.
5. There is a central duct running through the entire length of the testes. Each lobule is connected to the central duct by very minute ductules through which the ripe germ cells are passed down into the vas deferens.
6. The wall of the central duct consists of a single tier of epithelial cells surrounded by a very thin muscle layer.
7. The vas deferens is nearly a straight tube running latero-ventrally to the alimentary canal and extending from the beginning of the eighth abdominal segment upto the middle of the ninth abdominal segment. It is divided morphologically into two regions, an anterior thin region and a posterior enlarged region.
8. Histologically also the vas deferens is divided into two regions which correspond to the morphological divisions. The wall of the anterior region does not show a muscle layer which is, however, distinctly present in the posterior region.
9. The epithelial layer of the vas deferens is not lined internally by chitinous intima.
10. The two vasa deferentia open into a large saccular structure, the sperm sac situated in the middle of the ninth abdominal segment.
11. The histological structure of the sperm sac is similar to that of the last portion of the vas deferens except with the differences that the musculature is strongly developed in the sac and the epithelial cells are glandular.
12. The wall of the sperm sac is not lined internally by chitinous intima.
13. The sperm sac opens out ventrally to the exterior through a minute ejaculatory duct which is in the form of a deep cup-like pit.
14. The male genital opening is a longitudinal slit situated inbetween the bases of a pair of pear-shaped coxites in the middle of ninth abdominal segment on the ventral side.
15. The anal appendages consist of a pair of supra-anal appendages and an infra-anal appendage which is present only in the males.
16. There are seen teeth-like protruberances arranged in rows, on the outer ventro-lateral side of each supra-anal appendage. These have not been reported so far and are secondary sexual in nature.
17. The cup-like chitinous structure present dorsal to the male gonopore, is homologous to the rudimentary 'true penis' of the dragonflies.
18. The secondary copulatory apparatus consists of a genital fossa, a pair of genital lobes, an anterior lamina, a posterior lamina, lateral supporting framework, a penis sheath, a pair of hamules, a penis vesicle and a penis.
19. The shape of the anterior lamina, penis sheath and hamules is characteristic of the species studied.

20. The penis is a highly complex organ situated in the region of the second abdominal segment, anterior to the penis vesicle. It is distinctly three segmented, the third segment being bent over the first.
21. The orifice of the penis is situated near the distal end of the third segment on the dorsal side.

KEY TO LETTERING

A : Antero-lateral sclerite; B : Lateral arms of the penis sheath; C : Triangular chitinized sclerite on the dorsal side; CL : Cleft; CX : Coxites; ED : Ejaculatory duct; EP : Epithelium; f : Folds; G : Germ cells; H : Horizontal rod; HA : Hamule; HK : Hook-like structure; L : Lobule; LB : Lateral bar; MS : Musculature; O : Orifice; N : Latero-ventral sclerite; P : Postero-medial sclerite; PS : Penis sheath; RP : Rudimentary penis; R : Lobe-like sclerite; S : Sclerites with irregularly indented margin; SF : Supporting framework; SS : Sperm sac; TS : Testis; T : Lateral sclerites; U : Laterally compressed sclerites; VD : Vas deferens; VP : Ventral process; X : Chitinous process
h : Hairs.

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STUDIES ON LUNGWORMS (METASTRONGYLIDAE LEIPER,
1908) PARASITISING INDIAN LIVESTOCK

II. OBSERVATIONS ON NATURAL INFESTATIONS WITH SPECIES
OF *Dictyocaulus* RAILLIET AND HENRY, 1907 AND
VARESTRONGYLUS BHALERAO, 1932 IN THE SHEEP
OF HILLS IN UTTAR PRADESH

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In the examination of sixty two sheep lungs at the slaughter house at Nainital during November 1959 the incidence of the thread lungworm, *Dictyocaulus filaria* (Rudolphi, 1809) Railliet and Henry, 1907, *D. unequalis* Bhalerao, 1932 in the bronchi and bronchioles and *Varestrongylus pneumonicus* Bhalerao, 1932—the hair lungworm, in the bronchi, bronchioles and parenchyma of the lung was encountered, the latter species was invariably seen with characteristic lesions. The sheep, all in good condition and slaughtered for meat, were of two breeds—the Hill sheep (*Ovis nabhura*) of which seven were available and the Tibetan sheep (*Ovis hodgsoni*) of which fifty five were examined post-mortem.

Previous records of sheep infestations in the hilly tracts of this country with these species are those of Bhalerao (1932, 1934 and 1935), Baylis (1936), Srivastava (1945) and Mohan (1948) who, however, reporting the common occurrence of *D. filaria* and pulmonary changes associated with it in the plains sheep in Calcutta slaughter-houses, observed its greater occurrence in Hill sheep at Darjeeling.

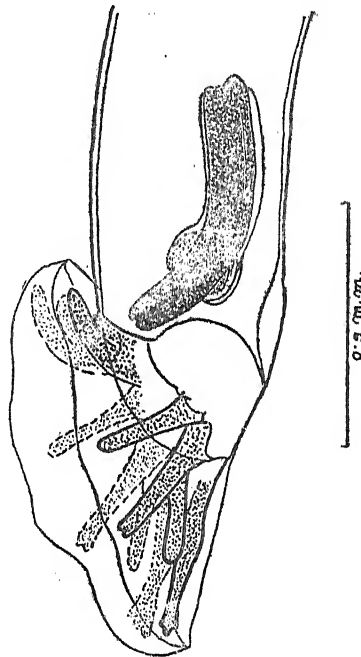
The importance of this group of roundworms in relation to the respiratory diseases of sheep has long been recognized all over the world and of late important work on some of the different aspects of parasitic broncho-pneumonia (including immunological responses) which, at times particularly in young and growing flock, may be associated with high degree of mortality and morbidity, is being done.

The present communication, besides enumerating briefly the main features in the biology of these species from the material available for examination, supplements a few essential anatomical features of these worms, their host-parasite relationships and the diagnostic characters of their first stage larvae which, for a correct assessment of the pathogenic significance, afford the only reliable aid. In addition, *in vitro* development of *D. filaria* has been studied.

Dictyocaulus filaria (Rudolphi, 1809)

Railliet and Henry, 1907.

Before Bhalerao (1932) described his new species *D. unequalis*, this lungworm was the only representative of the genus known from India. This new species, described from three female and one male specimens collected from large bronchi of Tibetan sheep, was differentiated by Bhalerao from *D. filaria* in having the spicules smaller and branches of the ventral rays unequal instead of equal. Baylis (1936), in his key to the species of *Dictyocaulus*, separates *D. unequalis* on the spicular length being slightly less than 0.3 mm while in *D. filaria* the spicules are stated to be between 0.3 and 0.6 mm. long. Descriptions of bursa no doubt exist in the compilation of Baylis and Yorke and Maplestone (1926) but it is to Gerichter (1951) that we owe a detailed account of the anatomy of *D. filaria*. This author has correctly stated that the ventrals are united at their proximal third while the ventro-ventrals are considerably shorter than the latero-ventrals and the two short spicules are stout, identical and 0.49–0.6 mm. in length. Thus, this unequal feature of the ventral rays characterises also *D. filaria* (Text Fig. I) and



Text Fig. I.

Lateral view of bursa (*D. filaria*).

consequently leaves us with only one of two characters on which Bhalerao had placed reliance in separating his species. In the large number of specimens collected during the present study the spicular length ranged from 0.31–0.53 mm. and *D. unequalis* can, therefore, no longer be retained on what now appears as only a slight variation in the matter of the measurements and this species is herein dropped. Our specimens agree essentially in all respects with the description given by Gerichter including the bursa in which the gubernaculum is described as oval in shape, spongy and transparent measuring 63 μ . This structure has, however, been found to be 65 μ –110 μ long.

The adult worms, lying in masses of exudate inside the bronchi and bronchioles and often incriminated with parasitic bronchitis, are milky white in colour with the males 4·7 - 7·8 cm. and the females 4·8 - 10·0 cm. in length. In the histological study of the lung tissue harbouring the adult worms, a fourth-stage larva in a respiratory bronchiole and younger juvenile in smaller bronchiole have been cut serially. The detection of such stages in naturally infested material affords a valuable clue to the understanding of the biology of the parasite in relation to its host-parasite relationship. These, as also the post-embryonic development in this case leading to its infective larvae studied during first half of November, 1958 in tap-water are briefly incorporated herein.

HISTOLOGICAL STUDY

These worms occur generally in the larger bronchioles in thick mucus with fibrin and number of lymphoid cells—the apical region of the affected lobes showing lesions. The bronchiolar mucous membrane consequently exhibits a thickening due to the enlargement of its mucus-secreting cells and at places a greater part of the epithelial lining gets denuded, the separated epithelial cells lying in the lumen (Plate I a). In the peri-bronchiolar region a heavy infiltration with lymphocytes

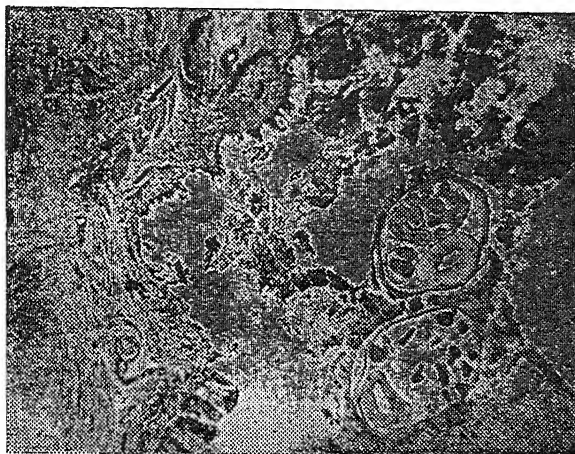


Plate I.

- (a) Photomicrograph from a section with adult *D. filaria* cut inside large bronchiole showing thickened and denuded portions of mucous membrane. 60 x

and polymorpho-nuclear leucocytes is encountered, the mucous glands being enlarged with surrounding blood vessels congested. A hypertrophy of the muscular coat and an increase in serosal membrane can also be observed. The finer bronchioles, connected with the affected larger bronchiole, also exhibit a congestion of its mucous membrane along with a marked infiltration with lymphocytes, polymorphs and also a few macrophages around them. The lymphoid tissue gets markedly hypertrophied with surrounding veins congested, thickened and some amount of haemorrhage is noticeable at places. The alveolar septa are thickened with a number of round cells and collapse of alveoli, filled with exudate, is visible. It is in such areas that a patchy consolidation is detected.

The fourth-stage larva, also inside a respiratory bronchiole, (Plate I b) exhibits an infiltration with lymphocytes in and around the bronchiolar wall. Similarly, the younger juvenile, inside a smaller bronchiole and possibly on its way to the larger bronchiole, in sections of this region and in the tissue around, exhibits clearly a destruction of the bronchiolar wall (Plate I c).

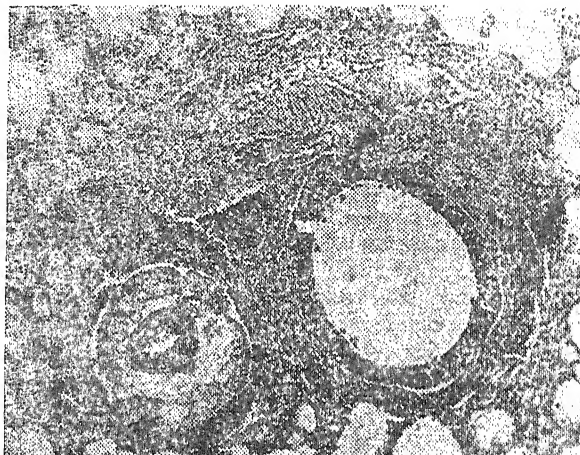
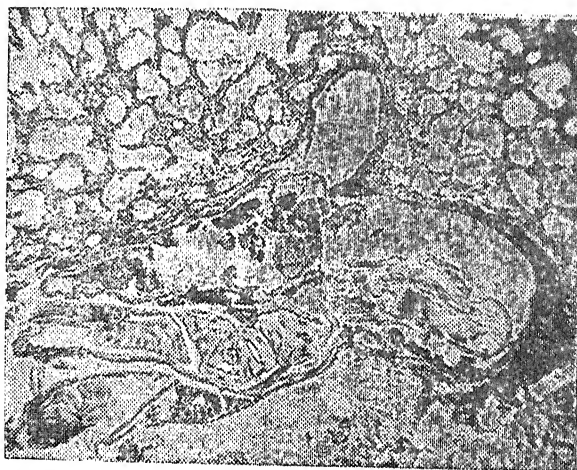


Plate I

- (b) Photomicrograph from a section showing fourth-stage larva of *D. filaria* cut inside a respiratory bronchiole with lymphocytic infiltration around the latter. 120 x



- (c) Photomicrograph from a section showing a younger juvenile cut inside a smaller bronchiole with destruction of its wall. 50 x

Varestrongylus pneumonicus Bhalerao, 1932

Bhalerao created this metastrongylid genus for the hair lungworm which he obtained from the large as well as smaller bronchi of both sheep and goats at Muktesar with *V. pneumonicus* as its type species. Sarwar (1944), without giving any description, proposed a new species of this genus as *V. capricola* which, however,

was later described by him in 1947 with a number of figures. Dougherty and Goble (1946) in their preliminary note have listed under this genus, the species, *V. pneumonicus*, *V. schulzi* (Boev and Vol'f, 1938) and *V. sagittatus* (Mueller, 1890) Dougherty, 1945—the latter only tentatively, this change in the conception of the genus resulting by regarding *Bicaulus* Schultz and Boev, 1940 as synonym of *Varestrongylus*. Of the two new lungworms from the small bronchioles and lung tissue of sheep and goats from China, *V. sinicus* Dikmans (1945), which was differentiated from *V. pneumonicus* in the form of its gubernaculum, had also been listed by Dougherty and Goble as a synonym of *V. schulzi*. According to Dougherty (1945), three species of the genus *Pneumstrongylus* Monnig 1932, needing transfer to *Varestrongylus*, were *P. sagittatus*, *P. alpenae* Dikmans (1935) and *P. caprioli* (Strop and Schmid, 1938). Dikmans paper was not incorporated by Sarwar nor by Dougherty (1951). The latter has, however, critically examined Sarwar's description of *V. capricola* and the conclusion that "Sarwar had not made out the true gubernacular structure in his material" is quite correct. Recently Sarwar (1955), after giving a number of figures under *V. pneumonicus* and *V. capricola*, reported that the former is a very common parasite of sheep and goats throughout Himalayas while the latter occurs as a rare species side by side with the former. No fresh details have been advanced by Sarwar in this paper in answer to Dougherty's criticisms. Boev (1957) has transferred the two species of *Varestrongylus*, *V. capricola* and *V. alces*, to the genus *Bicaulus*.

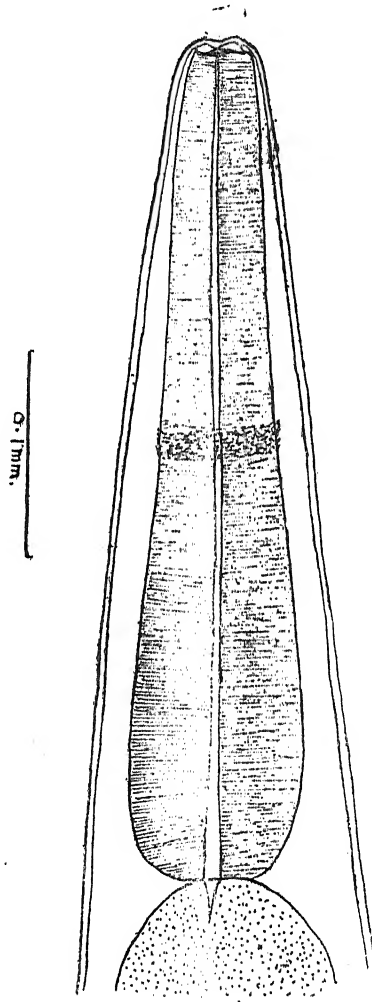
The material of hair lungworm collected at Nainital belongs to the genus *Varestrongylus* which has its outstanding generic characters in the structures of its gubernaculum in the male, the vulvar flap or sheath—designated as valve by Bhalariao in female and its study has necessitated a detailed comparison with the account of the two species given by Bhalariao and Sarwar particularly with reference to external genitalia and the accessory pieces—gubernaculum, telamon and the vulvar flap. This discrepancy in the two accounts and incomplete making-out of these structures have resulted in some degree of confusion and, therefore, a re-study of these parts from the present material is called for.

The following lines also include observations on the biology of this parasite inside the lung tissue, bronchiolar and alveolar, and the attendant changes.

This species appears to enjoy a wide distribution in the Hills both in sheep and goats and has been collected from the bronchi and bronchioles. A collection of adult worms is greatly facilitated during examination if the greyish-white and irregularly-shaped but usually symmetrical lesions are incised. It is this site as also the connected bronchi and bronchioles that invariably yield some specimens. The lesions somewhat diffuse, from 2.5–5 cm. in areas and slightly raised from the general pulmonary surface, occur mostly on the dorsal aspects in the posterior regions of the diaphragmatic lobes extending to costal and mediastinal surfaces and are seen also on other lobes as well. It is their larger size and the diffuse nature that distinguish these from those due to the other hair-lungworm, *Protstrongylus rufescens* (Leuckart, 1865) Kamensky, 1905, encountered in sheep in regions of the plains in which these have been seen to be from 0.5–1.5 cm. in diameter and occurring near the middle of the diaphragmatic lobes on the dorsal aspect but usually symmetrical in both the lungs. The adult specimens, often present mixed with *D. filaria* in the bronchi and bronchioles, generally inhabit in large numbers the smaller bronchioles connected with the lesions.

Adult worms are thin, of an orange-yellow colour with a thick, opaque and slimy pus around them which makes their detection somewhat difficult. The males are 1.4–2.0 cm. long and 0.17–0.2 mm. in thickness while the female worms

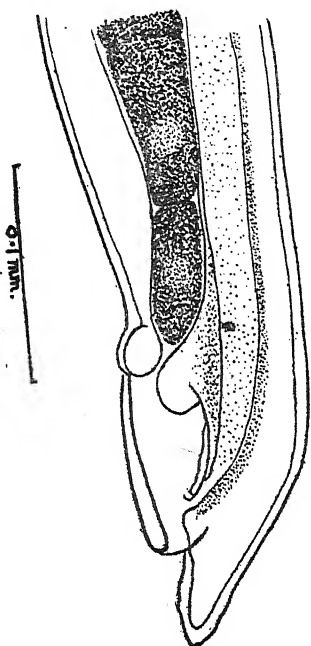
measure 2.53 - 3.1 cm. in length and 0.16 - 0.18 mm. in thickness. Mouth has around it the four lips and the club-shaped oesophagus (Text Fig. II) is 0.31 - 0.36 mm. long in males and 0.36 - 0.43 mm. in females, in which the tail is 0.051 - 0.06 mm. and the distance between anus and vulva, with two prominent lips nearly 0.024 mm. in size, is 0.06 - 0.08 mm. The cuticular flap or sheath, originating in the region of the anterior vulvar lip, 0.088 - 0.1 mm. in length is nearly tumbler-shaped and encloses the anal opening too. (Text Fig. III). This cuticular structure, the 'valve' of Bhale-rao, according to him measures 0.065 to 0.08 mm. in length and has been described as being inserted immediately anterior to vulva and disposed posteriorly. Sarwar simply mentions that the vulvar flaps are vestigial only. The characters of the



Text Fig. II.

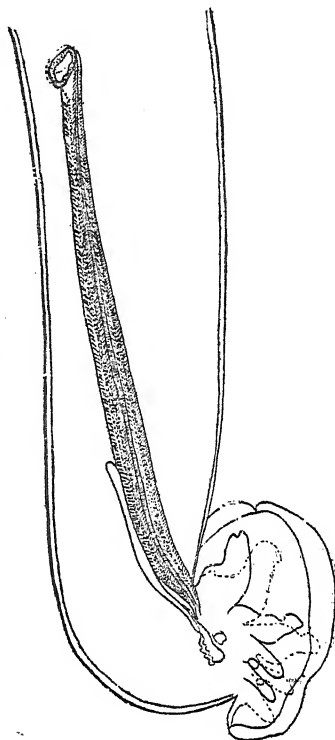
Anterior end (*V. pneumonicus*). . .

bursa, its rays and the spicules (Text Fig. IV) all agree with the description given by Bhalerao. The gubernaculum in *V. pneumonicus*, 0.138–0.165 mm. in length, has been described by Bhalerao as almost spindle-shaped with a tapering anterior end and a somewhat blunt posterior end which has two pairs of lateral processes, one situated at the extreme posterior end is very short and the other, which is about three times as long as the first, is situated at a distance of about 0.02 mm. in front of the former." Two chitinoid structures, measuring 0.026–0.027 mm. in length and with a swelling at their base have been described by him as telamon.



Text Fig. III.

Posterior end of female to
show vulvar flap (*V. pneumonicus*).

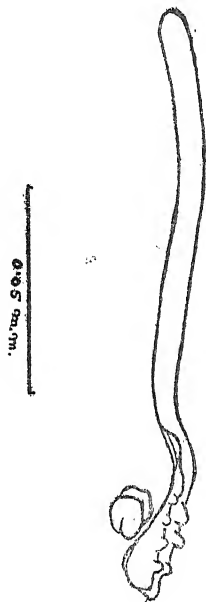


Text Fig. IV.

Lateral view of bursa
(*V. pneumonicus*).

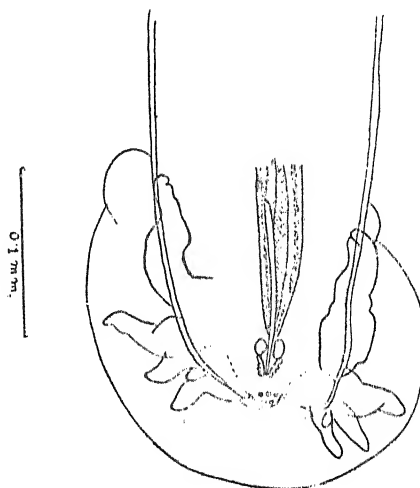
A careful study of the bursa from the specimens at our disposal shows that, unlike *P. rufescens*, the gubernaculum (Text Fig. V) is constituted by a common fused corpus, 0.084–0.092 mm. in length and the two crura, 0.036–0.04 mm. long and with four dentations on their outer edges, which have been mistaken by Sarwar for telamon—the latter has been described by him as consisting of two yellowish chitinous pieces each measuring about 0.025 mm. in length and having indentation on their outer edge. Bhalerao's two pairs of lateral processes are probably the two of the four indentations on each side of the crura but this author has, however, correctly identified the telamon which measures 0.015–0.016 mm. in size. In respect of the dorsal ray, the terminal digitations are five according to Bhalerao while Sarwar

gives the number for his species as apparently six. In our specimens there are five projections only (Text. Fig. VI). The ventral rays in *V. capricola* have been described as undivided by Sarwar (1947) but these, according to Bhalerao and in our specimens, are divided at their extremities.



Text Fig. V.

Gubernaculum and telamon
(*V. pneumonicus*).



Text Fig. VI.

Dorsal view of bursa
(*V. pneumonicus*).

We agree with the view expressed by Dougherty (1951) that the description and the figures given by Sarwar are very schematic. He has reported at one place absence of spindle-shaped gubernaculum but surprisingly enough this type of structure has been illustrated by him in his later paper of 1955. From all these discrepancies in the accounts it appears without doubt that Sarwar was really dealing with specimens of *V. pneumonicus* alone and his species evidently cannot be maintained and is herein dropped.

The genus *Varestrongylus* resembles some of the genera like *Protostrongylus* but can be distinguished on account of absence of the capitulum, the fused character of the corpus and the dentigerous crura—the three elements of the gubernaculum, and the presence of the vulvar flap or sheath extending to the anus. *Protostrongylus* has in the three elements of its gubernaculum a distinct capitulum, two corpus and two crura and additionally the chitinous arc, a valvar flap being absent.

The generic diagnosis proposed by Bhalerao (1932) needs slight amendment to include the distinctive feature of its gubernaculum by adding the following :—

"Gubernaculum consisting of a fused corpus carrying two crura which have indentations."

HISTOLOGICAL STUDY

From a large number of serially cut stained section of affected lung tissue it has been possible to study the location of adult worms, masses of eggs in different stages of segmentation and the first-stage larvae in bronchioles and alveolar spaces with the attendant damage, the cellular infiltration and the resulting changes in the lung tissue which resemble those associated with *P. rufescens* infection in local sheep already dealt with in brief elsewhere (Bhatia, 1960). The damage to the lungs in both these species is caused by the biology of the adults, their first-stage larvae and the developmental stages that had reached the pulmonary tissues from the circulatory system. Li (1946) gave an elaborate account of the histopathology of small lungworm infection (*Protostrongylus* sp.) in sheep and goats in North-West China describing the lesions, both grossly and microscopically. No photomicrographs had, however, been appended.

The numerous sections studied give a clear picture of the tissue reactions caused by the adult worms, the egg masses and the larvae. These agree in full with the observations recorded by Li. A brief illustrated account of the salient features is herein attempted. Histological study of the lesions, as a whole, exhibits important features like thickening of the inter-lobular septa, muscular hypertrophy of the bronchiolar walls and thickening of the blood vessels, inter-alveolar tissue and pleura (Plate I,d). The adult females, when occurring in the alveolar tissue of the

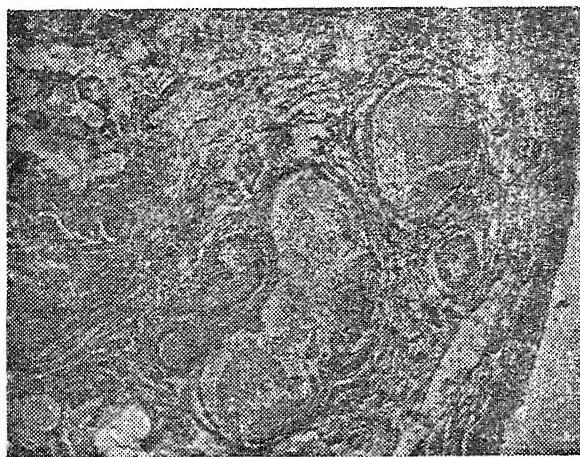


Plate I

(d) Photomicrograph from a lung section in *V. pneumonicus* infection showing thickening of inter-lobular septa, blood-vessels and pleura. 100 x

parenchyma presumably for egg laying, exhibit destruction of the alveolar walls, increased mucus production and haemorrhage as a result of mechanical irritation (Plate I,e). Egg masses, lying diffuse in the alveoli or outside them, mostly do not



Plate I

(e) Photomicrograph from a section showing adult female of *V. pneumonicus* cut in alveolar tissue with destruction of its wall and haemorrhage. 50 x

show any cellular changes (Plate II, a) but those in different segmentation and embryonal stages of development have around them an infiltration with lymphocytes and macrophages, some of the eggs in such masses being attacked by

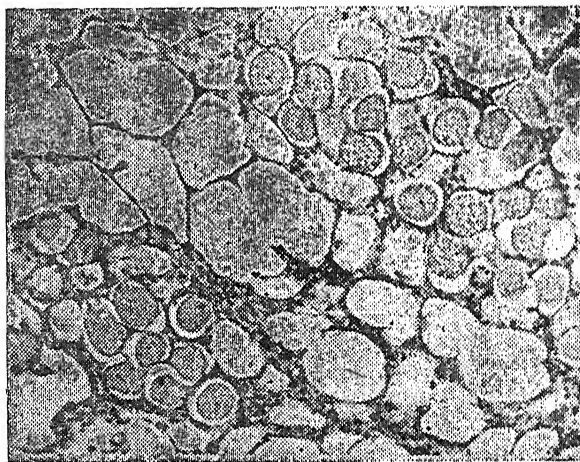


Plate II.

(a) Photomicrograph from a section showing an egg mass of *V. pneumonicus* in alveolar part with no visible reaction around. 210 x

macrophages and a few giant cells (Plate II, b). Eggs have also, though in very small numbers, been seen to lie in finer bronchioles. Larval masses, studied from different areas of lung tissue, lie in some cases near and around the respiratory

and finer bronchioles close to the destroyed alveolar tissue and the striking changes depicted relate to the broken alveolar walls, hyperplasia of some of the lining

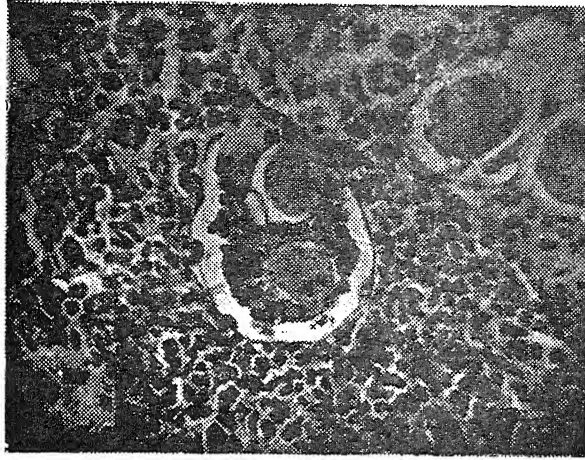
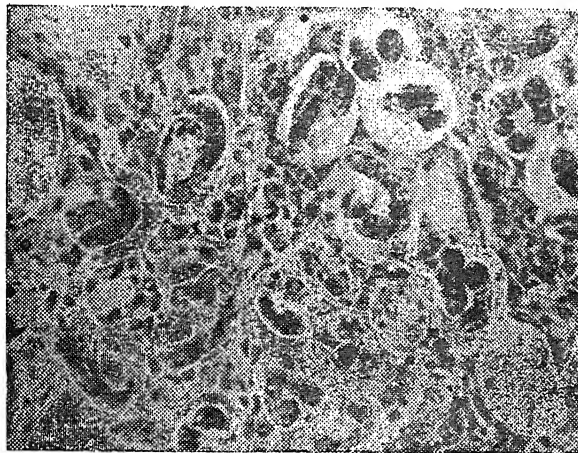


Plate II

(b) Photomicrograph from a section showing a few of the eggs of *V. pneumonicus* attacked by macrophages and a few giant cells. 400 x

cells, haemorrhage and in the reaction one can observe large number of lymphocytes around them (Plate II, c). A small number of macrophages with a few giant



(c) Photomicrograph from a section showing a mass of first stage larvae of *V. pneumonicus* with reaction around. 400 x

cells can also be observed attacking some of the larvae in the mass with pseudo-tubercle developing around them (Plate II, d).

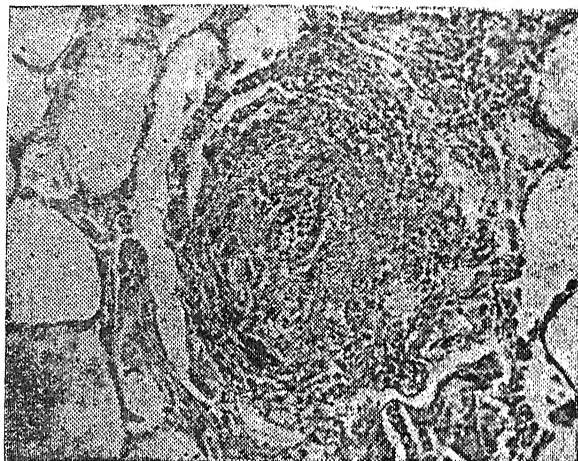
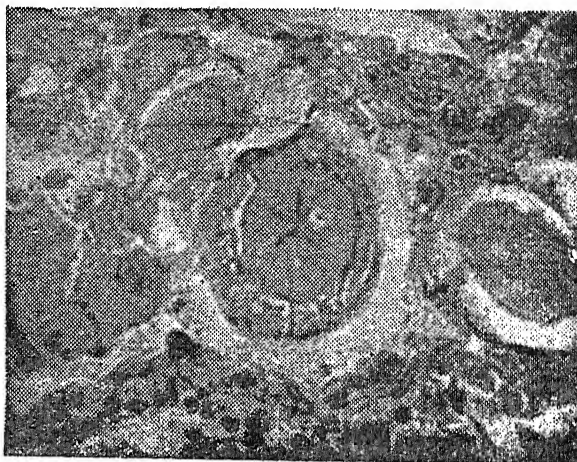


Plate II

(d) Photomicrograph from a section showing a first-stage larva of *V. pneumonicus* in a pseudo-tubercle. 210 x

Some of the sections also show the developmental stages received from the circulatory system prior to the formation of the adult worms, the former, in such cases, seen in the lung tissue show around them evidence of haemorrhage and cellular reaction of the type already described for the first-stage larvae (Plate II, e). No eosinophilic infiltration was detectable in any of the reactions.

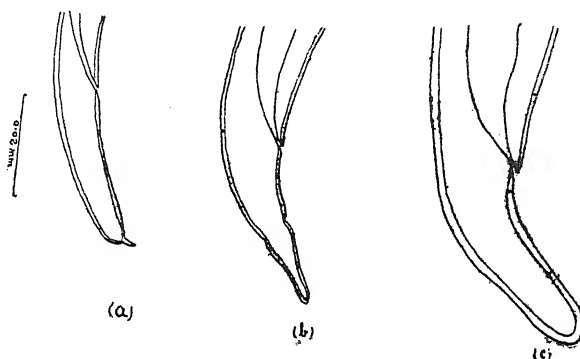


(e) Photomicrograph from a section showing the fourth-stage larva of *V. pneumonicus* (cut in oesophageal region) in alveolar tissue showing reaction around. 620 x

Further histological study of the lesions in *P. rufescens* infections in local sheep has shown that comparatively in this specific infection (*V. pneumonicus*) egg masses, as already pointed, are not so diffuse but the number in one mass is small and the eosinophiles, though present in small numbers, are predominated by lymphocytes. The changes due to hypertrophy and thickening etc., dealt with above, are similar for both the species.

FIRST-STAGE LARVAE RECOVERED FROM FAECES

In *D. filaria* the larva, measuring from 0.5–0.54 mm., has the characteristic small cuticular knob at its anterior end and the simple tail has a blunt ending (Text Fig. VII, c). The larva in *V. pneumonicus*, measuring 0.22–0.27 mm. in length, has the oesophageal and tail length 85–92 μ and 16–20 μ respectively. Its most distinctive feature lies in its straight tail ending in a short terminal spine, which is ventrally situated (Text Fig. VII, a). In *P. rufescens*, the larval length is 0.31–0.33 mm. and it is the wavy character of its tail showing curvature at two

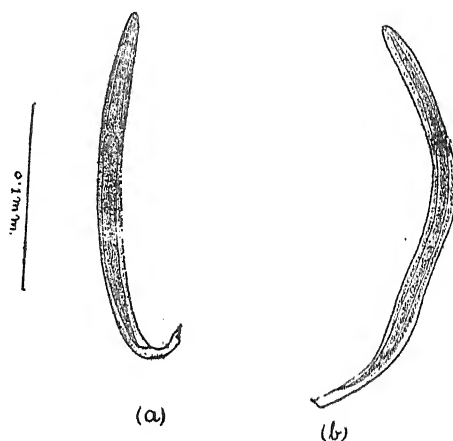


Text Fig. VII.

- (a) Tail of first stage larva of *V. pneumonicus*.
- (b) Tail of first-stage larva of *P. rufescens*.
- (c) Tail of first-stage larva of *D. filaria*.

points (Text Fig. VII, b) that is the distinguishing character. Some slight increase in the total larval length in the specimens collected from the faecal material is observed from those either recovered from the lesion or obtained from culture work which was attempted for *P. rufescens* (already dealt with elsewhere). In *V. pneumonicus*, the developing eggs and the first stage larvae present in the fixed lesions were also studied, the former measuring 68–80 $\mu \times$ 44–52 μ and the latter 0.21–0.25 mm. in length (Text Fig. VIII). The larval oesophagus was 84–90 μ , the tail measured 16–20 μ in length and the terminal spine was slightly longer

and some-what wavy (Fig. VIII, a) than that in the larva recovered from the faeces (fig. VIII, b).

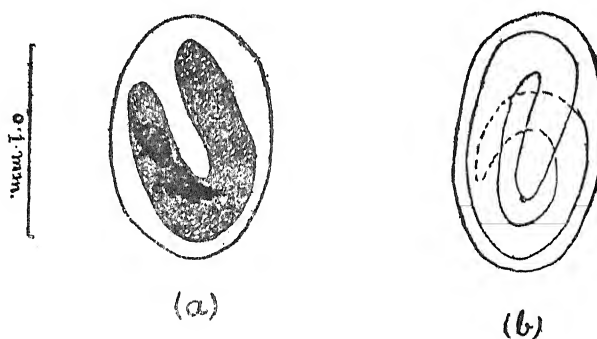


Text Fig. VIII

First-stage larva of *V. pneumonicus*,
 (a) recovered from lesion (fixed material).
 (b) recovered from faeces (fixed material).

POST-EMBRYONIC DEVELOPMENT IN *D. FILARIA*

Freshly collected females, left in tapwater, deposited embryonated eggs—the embryo inside consisting of a dark and nearly massive vermiform embryonic structure measured $120-136 \mu \times 68-88 \mu$ (Text Fig. IX, a). The culture work was carried



Text Fig. IX.

Embryonated eggs of *D. filaria*,

- (a) with dark massive vermiform embryo.
- (b) with completely formed larva occupying entire space inside shell, just before hatching.

out in laboratory with temperature and humidity conditions at 68–78°F and 58–68 % respectively. Most of the eggs by the following day had developed the fully formed coiled stage filling the entire space within the egg shell (Text Fig. IX, b). Regular hatching started after the third day. The first-stage larva (Text Fig. X, a), slender, of dark colour evidently because of the food granules contained in the intestinal wall and with a characteristic cuticular knob at the anterior end, measured from 0.47–0.53 mm. with the oesophagus 90 μ long and the simple straight tail with a blunt end measuring 25 μ in length. These larvae remained active for about twenty-four hours but later became lethargic and gradually motionless. In about seven days, after the eggs had been laid, the first ecdysis took place and the second stage larvae (Text Fig. X, b), ensheathed with the old cuticle, measured 0.5–0.55 mm. with the oesophagus and tail 100 μ and 26 μ long respectively. After a short span of activity these larvae entered into the lethargic state and finally became gradually motionless and it was thus nearly



Text Fig. X.

Developmental larval stages of *D. filaria*.

- (a) first-stage,
- (b) second-stage,
- (c) third (pre-infective) stage,
- (d) third (infective) stage.

twelve or thirteen days after the egg laying that all the larvae had completed the second ecdysis and reached the third stage (Text Fig. X, c), designated as the pre-infective stage by Gerichter (1951), within the two sheaths. These larvae measured 0.54–0.56 mm. in length with the oesophagus 100 μ and the tail 28 μ long, the two sheaths beyond the tip of the tail being 2–3 μ long. The characteristic cuticular knob at the oral end and the dark granular stored food reserves in the alimentary tract characterised all the three developmental stages. This culture

was subsequently fixed in hot 70% alcohol for later study. The numerous specimens, after clearing, were of three types :—(i) with the two sheaths lying closely adhered to the larval skin, (ii) with the outer of the two sheaths widely separated on one side and (iii) with only one sheath, there being absolutely no change in the body measurements in all the three kinds. The first category belongs to the pre-infective group, the second is in the process of losing the outer of the two sheaths and the third is the infective stage larva, (Text Fig. X, d) the period taken by the last to develop was not observed in this culture work.

The present study lends support to the view that inspite of heavy infestations and characteristic lesions over a greater or smaller area of pulmonary tissue, the animals appeared to have tolerated the infections. This may have been due to age resistance or some immunological mechanism. All such cases, therefore, may have a major role in acting as carriers and reservoirs of infection or latent cases and also in greatly contaminating the limited land available for feeding and grazing. In order to protect the younger stock from the serious damage of the type associated with mortality and morbidity in consequence of secondary pathogenic infections it may be more advantageous to concentrate on animal husbandry practices that are exclusively based and practised on scientific lines particularly when a programme of improving our sheep from cross-breeding with certain imported breeds is being undertaken.

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ON TWO NEW AVIAN CESTODES BELONGING TO THE SUBFAMILY
HYMENOLEPIDINAE PERRIER, 1897 FROM DELHI STATE

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The material, which forms the basis of this paper, was obtained from the birds shot in Timarpur Delhi. The author is greatly indebted to the Council of Scientific & Industrial Research for the financial assistance in course of the investigations. All measurements unless otherwise mentioned are given in mm.

Family HYMENOLEPIDIDAE Railliet & Henry, 1909

Subfamily HYMENOLEPIDINAE Perrier, 1897

HYMENOLEPIS Weinland, 1858

Hymenolepis jasuta n. sp.

Host : *Coturnix coturnix* (Linnaeus).

Maximum length is 100 & the greatest breadth 0.29 - 0.316 (mature segments), and 0.475 (gravid segments). Scolex is 0.305 long and 0.22 broad. Rostellum is 0.075 in diameter. Rostellar sac measures 0.275×0.088 extending below the lower margin of the suckers. Rostellar hooks number ten, measuring 0.103 in length. Suckers, almost spherical, measure 0.095 - 0.103 in diameter. Genital pores are unilateral, usually located slightly anterior to middle or sometimes near

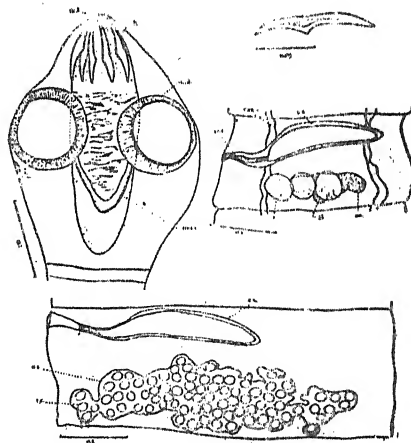


Plate I—*Hymenolepis jasuta* n. sp.

- A. Scolex.
- B. Rostellar hook.
- C. Mature segment.
- D. Gravid segment.

anterior one fourth part of the proglottis margin. Cirrus sac measures $0.225 - 0.246 \times 0.037 - 0.047$ (mature segments) and 0.314×0.047 (gravid segments),

crossing ventral longitudinal excretory vessel, and extending upto aporal ventral longitudinal excretory vessel and even crossing it in some cases. There are three testes arranged usually in a transverse row in posterior half of the segment in between the longitudinal excretory vessels ; occasionally they show some variation in their position as listed below :—

Arrangement of the testes

| Testes pattern | Disposition |
|--------------------------|-------------|
| 1. Straight line (usual) | - * * * |
| 2. Variant (triangular) | - * * * |
| 3. „ („) | - * * * |
| 4. „ („) | - * * * |

Key to the above :—

- ... Genital pore.

* ... Testis

The diameter of testes varies 0.035 – 0.46. Ovary is in close association with two or all the three testes and is disposed transversely : its anterior margin is uniform while the posterior one is irregular with a few small lobes produced from that side. A small vitelline gland (0.02 – 0.022 diameter) is placed posterior to ovary. A small shell gland lies in between the ovary and the vitelline gland. Uterus is an irregularly lobed sac extending lateral to the excretory vessels and encloses large number of egg-capsules. The eggs and onchospheres measure 0.021 – 0.026 and 0.014 – 0.019 respectively.

The accompanying table I shows a number of species under *Hymenolepis* Weinland 1858 where the cirrus sac is fairly large and extends upto the aporal ventral longitudinal excretory vessel. The present form represents outstanding characters on account of very long size of the rostellar hooks, the absolute size and the relative extent of the cirrus sac together with the disposition of the genital organs (chiefly the extra-ordinary extension of the ovary having close association with all the testes.) It is, therefore, essential to create a new species for the reception of the present material.

Hymenolepis jerratta n. sp.

Host : *Erolia minuta minuta*.

The specimens of this material vary from 54.0-68.0- in length and 0.68 – 1.14 in breadth. The scolex is well developed structure measuring 0.283 in length and 0.4 in breadth and is bearing suckers on its anterior part and they measure 0.12 – 0.128 in diameter. The rostellum is very small, measuring 0.025 – 0.03 in diameter and a small portion 0.040 long protruding out of the scolex and carries 8 – 10 small

Table I

Genus—*Hymenolepis* Weinland, 1858

Species with cirrus sac reaching aporal ventral longitudinal excretory vessel

| Species | R. h. (no.) | R. h. (size in μ) | Cirrus sac (size in μ) | Cirrus sac* (extent) | Genital pore | Disposition of testes in a segment | Additional character |
|--|----------------|------------------------------|--------------------------------|-------------------------|--------------|--|-------------------------|
| <i>H. exigua</i> Yoshida 1910 | 10 | 30–50 | 300 | to aporal one | 1/3 ant. | - * * | |
| <i>H. falcata</i> Meggitt 1924 | 10–12 | 56–58 | | " | 1/4 ant. | - * * | |
| <i>H. felicia</i> Meggitt 1927 | 24 | 34–52 | 130–140 x 28–44 | " | 1/3–1/4 ant. | - * * | |
| <i>H. longicauda</i> Fuhrmann 1906 | ? | ? | ? | " | 1/3 ant. | - * * | |
| <i>H. microphs</i> Diesing 1850 | ? | 16 | 115–118 | " | anterior | - * * | |
| <i>H. multiglandularis</i> Baczynska 1914 | 10 | 28.6, 23–33 | ? | " | 1/3 ant. | - * * | |
| <i>H. octatentha</i> (Krabbe 1869) | 8 | 32–34 | ? | past aporal one | 1/6 ant. | - * * | |

*In relation to ventral long. exc. vessel.

Table 1 (contd.)

| Species | R. h. (no.) | R. h. (size in μ) | Cirrus sac (size in μ) | Cirrus sac* (extent) | Genital pore position on proglottis margin | Disposition of testes in a segment | Additional character |
|---|----------------|------------------------------|-------------------------------------|--|---|--|-------------------------|
| <i>H. pauciovata</i> Fuhrmann 1906 Megitt 1927 | 10 | 65-70 | 50-60 \times 10 | To aporal one or near | 1/2 | - * * * | |
| <i>H. rustia</i> Megitt 1926 | 0 | 0 | 200-200 | To aporal one | 1/2 | - * * | |
| <i>H. singularis</i> Baer 1932 | 10 | 61-62 | 70-76 \times 20 | To aporal one (by fig.) | ? | - * * | |
| <i>H. terrae-reginae</i> Jonston 1911 | ? | ? | ? | Near to aporal one | 1/4 ant. | - * * | sacc. acc. present. |
| <i>H. tubicirrosa</i> Baczynska 1914 | ? | ? | 680-153 | To aporal one | 1/2 or ant. | - * * | |
| <i>H. jesusula</i> n. sp. | 10 | 103 | 225-246 (mature) 314 (gravid) | To aporal one and even be- yond it. | ant. to mid- dle near 1/4 anterior also varying. | - * * * (Normal) | |

*In relation to ventral longitudinal excretory vessel.

rostellar hooks, measuring 0.003 - 0.0035. Each rostellar hook bears a very significant guard and its shaft is very small, joined to the handle abruptly. A rostellar sac (measuring 0.147 in length and 0.094 in breadth) is well developed with a

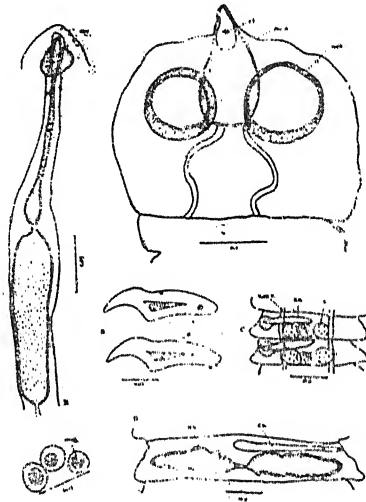


Plate II—*Hymenolepis jerrata* n. sp.

- A. Scolex.
- B. Rostellar hooks.
- C. Mature segments.
- D. Gravid segment.
- E. Cirrus sac (enlarged).
- F. Eggs.

voluminous area and a conical anterior extension; its posterior border is almost approaching the lower margin of the suckers but it does not extend below it. Genital pores are unilateral, situated a little anterior to the middle of the proglottid margin. Sometimes it is slightly shifted to the surface. Genital cloaca is well developed with thick walls and measures 0.068 - 0.09 in diameter. Cirrus sac is fairly long measuring 0.29 - 0.33 \times 0.04 - 0.048 in the mature segments and 0.50 - 0.61 \times 0.045 - 0.052 in the gravid segments. It extends upto the middle or a little over half of the proglottis breadth. Cirrus sac contains a large internal seminal vesicle measuring 0.25 \times 0.068 and connecting a prominent ductus ejaculatorius. Cirrus is simple without any spines. Three testes lie in a transverse row, almost touching the posterior border of the segment and are enclosed in between the two longitudinal excretory vessels. Occasionally one of the lateral testes may be partially lateral to the longitudinal excretory vessels. Their size varies from 0.07 - 0.12 in diameter. It has been observed in some of the mature segments that the middle testis is slightly smaller than the lateral ones.

In anterior segments where testes are fully developed, the ovary is a simple structure and is in close contact with the central testis. In the posterior segments, ovary shows distinct lobed character and measures 0.23 \times 0.11. A vitelline gland, measuring 0.07 - 0.09 in diameter, is located posterior to ovary.

The uterus is an irregularly transverse sac usually divided into two compartments with a narrow connection between them. The egg-capsules are filling up the uterine sacs and are almost spherical in form and measure 0.02-0.25 in diameter. The onchospheres measure 0.012-0.017 in diameter.

On comparison the present form approaches the following known species of the genus *Hymenolepis* Weinland, 1858, *H. columbae* (Zedar 1803), *H. corvi* Mayhew 1925, *H. fista* Meggitt 1933, *H. graülis* (Zedar 1803), *H. incognita* Meggitt 1927, *H. megalorchis* (Lühe 1898) *H. serrata* Fuhrmann 1906 and *H. serrata biramanica* Meggitt 1924. The possession of very small typical rostellar hooks, position of the genital pore, relative and the absolute size of the cirrus sac, a voluminus vesicula seminalis, the disposition of testes and ovary in the present form easily separate out the above mentioned species.

A new species, *Hymenolepis jerrata*, is therefore created for its reception.

SUMMARY

Hymenolepis jasuta n. sp.

Host : *Coturnix coturnix* (Linnaeus)

Maximum length 100. greatest breadth 0.316 (mature segments) and 0.475 (gravid segments). Scolex 0.305 long and 0.22 broad. Rostellum 0.275 in diameter. Rostellar sac 0.275 × 0.088 extending below the lower margin of the suckers. Rostellar hooks ten, 0.103 long. Suckers 0.095-0.103 in diameter. Genital pores unilateral. Cirrus sac 0.225-0.246 × 0.037-0.047 (mature segments) and 0.314 × 0.047 (gravid segments) extending upto aporal ventral longitudinal excretory vessel and even crossing it in some cases. Testes three, arranged normally in a transverse row, their variation is also studied. Ovary, in close association with two or all the three testes, disposed transversely. Vitelline gland 0.02-0.022 in diameter. Uterus irregularly lobed sac extending lateral to longitudinal excretory vessels. The eggs and onchospheres measure 0.021-0.026 and 0.014-0.019 in diameter respectively.

The present form represents outstanding characters on account of very long size of the rostellar hooks, the absolute size and the relative extent of the cirrus sac together with the disposition of the genital organs. It has also been compared with the allied forms possessing fairly large cirrus sac as listed in the accompanying table.

Hymenolepis jerrata n. sp.

Host : *Erolia minuta minuta*

Maximum length 68.0 and the greatest breadth 1.14. Scolex 0.283 long and 0.4 broad. Rostellum very small, 0.025-0.03. in diameter and a small portion 0.04 long protrudes out of scolex. Rastellar sac, 0.147 long and 0.094 broad, extending down below the lower level of the suckers. Rostellar hooks 8-10, 0.093-0.035 long. Genital pores unilateral. Genital cloaca well developed. Cirrus sac 0.29-0.33 × 0.04-0.048 in mature segments and 0.5-0.6 × 0.04-0.05 in gravid segments. A large internal seminal vesicle and a ductus ejaculatorias present. Testes three disposed in transverse row. Ovary approximately in the centre of the proglottis, in close contact with the central testis. Vittelline gland 0.07-0.09 in diameter. Ulterus an irregularly transverse sac usually divided

into two compartments with a narrow connection between them. The eggs and onchospheres measure 0.02–0.025 and 0.12–0.017 in diameter respectively.

The present form has been contrasted from the species of the genus *Hymenolepis* Weinland, 1858, which appear approaching it. The possession of very small typical rostellar hooks, the position of the genital pore, relative and absolute size of the cirrus sac, a voluminous internal vesicula seminalis, the disposition of the ovary and the testes in the present material clearly distinguish it as a new species.

ABREVIATIONS USED

| | |
|-----------|-----------------------------|
| cirr. | cirrus |
| c. s. | cirrus sac |
| e. c. | egg-capsules |
| i. v. s. | internal vesicula seminalis |
| ov. | ovary |
| onch. | onchospheres |
| r. h. | rostellar hooks |
| rost. | rostellum |
| rost. s. | rostellar sac |
| suck. | sucker |
| t. | testis |
| u. s. | uterine sac |
| v. ex. v. | ventral excretory vessel |
| v. gl. | vitelline gland |

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SEASONAL GROWTH OF OOCYTES OF *MYSTUS SEENGHALA* (SYKES)
AND *WALLAGONIA ATTU* (BLOCH) WITH AN INFERENCE
ABOUT THEIR SPAWNING HABITS

By

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[Received on 14th July, 1959]

INTRODUCTION

The maturity and the spawning periods of certain fishes have been studied by measurements of the dimensions of ova by Clark (1934), Hickling and Rutenberg (1936) and De Jong (1939). On the Indian fishes only a little work of this nature has been done. Among the notable Indian contributors the names of Arora (1951), Palikar and Karandikar (1952) and Prabhu (1956) may be mentioned. Investigations on the spawning period of a fish should normally require observations on the gonadic condition of the fish for at least a complete year yet it is found that most of these authors have confined their observations on studying the ovaries for a few months only in the year with the risk of rendering their accounts incomplete. In the present communication, the writer has, however, based his observations on the ovarian ova throughout the whole year, and by the measurements of these, has made an attempt to draw some inference about the spawning periods of the fishes.

MATERIAL AND METHOD

The material included two species of common Indian fishes, viz., *Mystus seenghala* and *Wallagonia attu*. They were collected from river Yamuna at Allahabad at the rate of two to three times in a week throughout one complete year. The size of fishes collected varied from 25 to 32 inches.

To have the measurements directly comparable, all the ovaries were fixed in 5% formalin. Clark (1934) in his studies on California sardine, however, used 10% formalin. In the present study 5% formalin proved to be a quite satisfactory preservative. Thin slices were cut from the middle portion of the ovaries, dehydrated, imbedded and sectioned about 7 to 8 μ thick. The measurements of the diameter of ova were recorded from these sections. Prabhu (1956) has also studied the diameter from the sections of the ovaries in his study on the spawning periodicities of *Macrones vittatus* and *Therapon jarbua*. The measurements were taken by the aid of a standardised ooculometer.

The author has, however, tried to investigate the range of the diameter of ova in the ovaries of these fishes in each month of the year. In spite of certain limitations he has adopted the following procedure:

The ooculometer was placed in a horizontal position and the diameters were measured parallel to its graduations in order to minimise the error due to the lack of symmetry in the oocytes due to preservation. From the sections of each ovary, two to three smallest and two to three largest oocytes were measured and recorded. Thus the measurements of the oocytes were recorded from about 8 ovaries in a month. Likewise the data for the entire year were obtained for each fish. From the data of each month of the year the smallest and the largest diameters were recorded for study. For the sake of convenience, the smallest and largest oocytes have been termed as "small" and "large" oocytes.

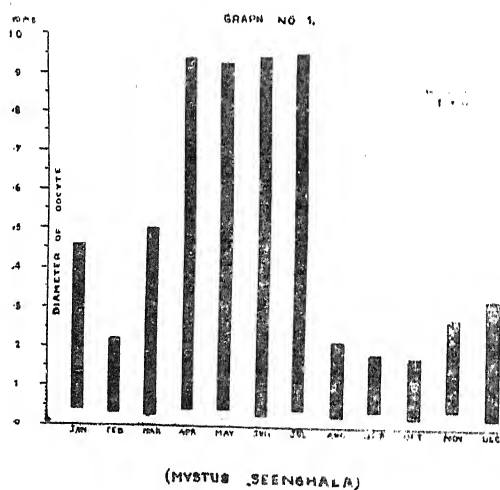
OBSERVATIONS

Mystus seenghala :—

The monthly data of the measurements of the small and large oocytes are as follows :—

| Months | Diameter of "Small" oocyte | Diameter of "Large" oocyte |
|-----------|-------------------------------|-------------------------------|
| January | ·042 mms. | ·462 mms. |
| February | ·035 " | ·220 " |
| March | ·028 " | ·504 " |
| April | ·042 " | ·945 " |
| May | ·012 " | 928 " |
| June | ·028 " | ·948 " |
| July | ·042 " | ·950 " |
| August | ·028 " | ·217 " |
| September | ·042 " | ·189 " |
| October | ·028 " | ·182 " |
| November | ·49 " | ·280 " |
| December | ·028 " | ·329 " |

These data have been plotted in a graph no. 1, to give an idea of the variations in the range of diameter of oocytes in the various months of a year.



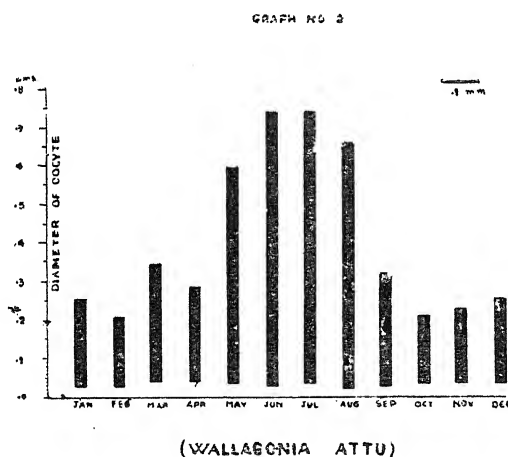
Graph No. 1. It shows the changes in the range of diameter of oocytes in the ovary of *Mystus seenghala* in the different months of a year.

Wallagonia attu :—

The monthly data of the diameter of "small" and "large" oocytes is furnished below :—

| Months | Diameter of "Small" oocyte | Diameter of "Large" oocyte |
|-----------|-------------------------------|-------------------------------|
| January | ·028 mms. | ·252 mms. |
| February | ·028 „ | ·210 „ |
| March | ·042 „ | ·343 „ |
| April | ·042 „ | ·287 „ |
| May | ·035 „ | ·595 „ |
| June | ·028 „ | ·742 „ |
| July | ·035 „ | ·745 „ |
| August | ·021 „ | ·665 „ |
| September | ·028 „ | ·322 „ |
| October | ·035 „ | ·210 „ |
| November | ·035 „ | ·224 „ |
| December | ·035 „ | ·259 „ |

The diameters of "small" and "large" oocytes in the various months of the year have been represented in the graph no. 2.



Graph No. 2. It depicts the variations in the range of diameter of oocytes in the ovary of *Wallagonia attu* in the various months of a year.

DISCUSSIONS

The examination of data and graphs of the diameter of the oocytes of both the fish genera *Mystus* and *Wallagonia* makes it apparent that there exists a certain parallelism between these two fishes in so far as the presence of small oocytes are concerned throughout the year and the response of large oocytes to the change of the months in a year is almost identical. The period prior to July in both fishes can be regarded as the period of gradual increase in the diameter of "Large" oocytes; and the period thereafter as a period of regression. The period up to July can thus be regarded as pre-spawning period as it shows the advancement in size of the "Large" oocytes. A close examination of the data and graph reveals that in the case of *Mystus*, the size of "Large" oocytes faces a sudden and sufficient fall in August attaining its almost pre-spawning size. In the case of *Wallagonia* also the diameter of "Large" oocytes faces a downfall in August, but here unlike *Mystus*, the pre-spawning size appears to be achieved a little later in the month of September. Thus it appears quite probable that in the case of *Mystus* the spawning lasts to a shorter duration than in the case of *Wallagonia*. In general the months of July and August can be inferred to be the spawning months. But any such hypothesis can only be tentative unless their spawning is subjected to careful investigation in their natural habitat. But as the peak of diameter of "Large" oocytes is reached only once in a year in both the fishes they can safely be inferred to be annual breeders. The present results on *Mystus Seenghala* differs from that on its allied species *Macrones (Mystus) vittatus* by Prabhu (1956) as he has inferred the latter to spawn during October and November in South India, while the former is inferred to spawn during July and August. This might be due to climatic differences between South India and Allahabad. The present study supports the work of Prabhu (1956) in the duration of spawning being short and its taking place only once in a year as he has reported *Macrones (Mystus) vittatus* also to spawn only once in a year and the duration of spawning to be short. Thus, these two species though differ in the months of their spawning, bear a close resemblance in their being annual breeders and the spawning lasting for short duration.

The study on *Wallagonia attu* supports the work of Ahmad (1944a). He has also reported this fish to spawn in July.

An important point which this study reveals from the perusal of the data of the diameter of "Large" oocytes, in both the fishes, is that the mature oocyte of *Mystus* is larger in size than that of *Wallagonia*. Evidently, the former produces eggs larger in size than those of the latter. Secondly, maturation of ova commences earlier in *Mystus* i.e. in the month of April, than that of *Wallagonia* where it starts from the month of May.

SUMMARY AND CONCLUSION

1. The ovary of both the fishes *Mystus seenghala* and *Wallagonia attu*, is never free from young oocytes throughout the year.
2. The "Larger" or mature oocytes attain their maximum size only once in a year in the month of July in both the fishes. Hence these fishes are inferred to be annual breeders.
3. *Mystus seenghala* appears to produce eggs larger in size than those of *Wallagonia attu*.

ACKNOWLEDGEMENT

The author is indebted to Dr. S. K. Dutta, D.Sc., under whose guidance the work was done in the research laboratory of the Zoology Department of Allahabad University.

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EFFECT OF VITAMIN B₁₂ AND ANTIBIOTICS ON HEMATOPOESIS IN THE CHICK

By

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INTRODUCTION

When the isolation of crystalline vitamin B₁₂ was announced by Rickes *et al.* (1) it was reported to be extremely effective in the treatment of Addisonian pernicious anaemia (2). These and other reports (3,4) have indicated that in the human, vitamin B₁₂ apparently stimulates the bone marrow to produce mature erythrocytes and myeloid leucocytes. Vitamin B₁₂ deficiency, was reported by Williams *et al.* (5) to have little or no effect on the hemoglobin level of chicks fed a diet supplemented with other B vitamins. Stern *et al.* (6) reported that hemoglobin and hematocrit and erythrocyte counts of vitamin B₁₂ deficient chicks were not affected by an uncomplicated deficiency of this vitamin although growth was severely retarded. Hsu *et al.* (7) demonstrated that hemoglobin and erythrocyte counts on vitamin B₁₂ supplemented embryo blood were significantly higher than on blood of vitamin B₁₂ deficient embryos during final stages of incubation. The present paper reports further observations regarding the effect of vitamin B₁₂ and antibiotics on hematopoiesis in the chick.

PROCEDURE

Day-old S. C. White Leghorn chicks were randomly distributed into 2 lots of 12 chicks each. The chicks were brooded in kerosine oil heated brooders with raised wire floors. They were fed a 31% protein, vitamin B₁₂ deficient diet, as shown in Table 1. Chicks in lot 1 received the basal diet while chicks in lot 2

TABLE 1
Composition of Vitamin B₁₂ deficient diet.

| Ingredients | Amounts % |
|--|----------------|
| Maize | 31.0 |
| Groundnut Cake | 65.0 |
| Mineral Supplement (ICI brand Mineral Mixture). | 2.0 |
| Limestone | 1.0 |
| Shark Liver Oil (1000 I. U. Vitamin A and 100 I. U. Vitamin D/gm). | 1.0 |
| Manganese Chloride | 8 gm./100 lbs. |
| Riboflavin | 5 mg./lb. |
| Thiamin | 3 mg./lb. |
| Calcium Pantothenate | 7 mg./lb. |
| Niacin | 20 mg./lb. |
| Pyridoxine | 3 mg./lb. |

received the basal diet supplemented with 3 p.p.m. Procaine penicillin. Six chicks from each lot were intramuscularly injected with 50 mcg. of vitamin B₁₂ weekly. At six weeks of age each lot was divided into 4 groups of 3 chicks each, thus forming 8 groups of 3 chicks each. To hemolyze the mature red cells, four groups received a single subcutaneous injection of phenylhydrazine HCl at the rate of 2 mg. per 100 gm. body weight.

Blood was drawn from the brachial vein, 1,3,5, and 7 days after the injection of phenylhydrazine HCl, using Potassium Oxalate as an anticoagulant. Body temperature, erythrocyte counts, hematocrit, hemoglobin and specific gravity determinations were made. Body weights were recorded on alternate weeks.

Erythrocyte counts were made by means of a standard Fein-Optic hematocytometer having improved double naubauer ruling. Hemoglobin was determined by means of a hemometer graduated on Hellige scale and the hematocrit by centrifugation at 2000 r. p.m. for 10 minutes and the volume of packed cells determined.

Specific gravities of whole blood were determined according to the Copper Sulfate method of Van Slyke *et al.* (8). Body temperature was taken by means of a 'Zeal' clinical thermometer graduate into °F by inserting it into the vent for 2 minutes at a depth of 3 cms. Each determination was made in duplicate.

RESULTS AND DISCUSSION

The results of the hematological study are presented in Tables 2, 3 and 4. Injection of phenylhydrazine HCl in the absence of vitamin B₁₂ resulted in a significantly lower blood content of hemoglobin, erythrocyte and hematocrit. The effect was less pronounced when both phenylhydrazine HCl and vitamin B₁₂ injected.

TABLE 2
Effect of vitamin B₁₂ and antibiotics on Hemoglobin regeneration in chicks.

| Serial no. | Treatment | | | Hemoglobin, gm%, days after injection of phenylhydrazine HCl | | | |
|---------------|---------------------------------|-----|--|---|------|------|--------|
| | Supplement to the basal diet | | Injection | 1 | 3 | 5 | 7 |
| 1 | None | ... | None ... | 7.8 | 7.8 | 7.8 | 8.2 |
| 2 | None | ... | Phenylhydrazine HCl | 4.5** | 5.5 | 6.6 | 8.3 |
| 3 | None | ... | Vitamin B ₁₂ and Phenylhydrazine HCl. | 5.0** | 7.5 | 8.3 | 9.1 |
| 4 | None | ... | Vitamin B ₁₂ ... | 9.8 | 10.0 | 10.0 | 10.1** |
| 5 | Penicillin | ... | None ... | 8.3 | 8.9 | 8.7 | 8.8 |
| 6 | Penicillin | ... | Phenylhydrazine HCl. | 4.4** | 6.3 | 6.6 | 8.5 |
| 7 | Penicillin | ... | Vitamin B ₁₂ and Phenylhydrazine HCl. | 4.9** | 6.9 | 8.6 | 9.1 |
| 8 | Penicillin | ... | Vitamin B ₁₂ ... | 10.2 | 10.2 | 10.2 | 10.4** |

** P < .01 compared to control groups 1 and 5.

TABLE 3

Effect of Vitamin B₁₂ and Antibiotics on Erythrocyte Count of chick blood.

| Serial no. | Treatment | | | | Erythrocyte counts, $\times 10^6/\text{cu. mm.}$, days after injection of phenylhydrazine HCl | | | |
|------------|--------------------------|-----|--|-----|--|------|--------|-------|
| | Supplement to basal diet | | Injection | | 1 | 3 | 5 | 7 |
| 1 | None | ... | None | ... | 1.86 | 1.85 | 1.94 | 2.00 |
| 2 | None | ... | Phenylhydrazine HCl. | ... | 1.01** | 1.34 | 1.62** | 1.92 |
| 3 | None | ... | Vitamin B ₁₂ and Phenylhydrazine HCl. | ... | 1.10** | 1.70 | 1.97 | 2.10 |
| 4 | None | ... | Vitamin B ₁₂ | ... | 2.12 | 2.20 | 2.10 | 2.19* |
| 5 | Penicillin | ... | None | ... | 1.93 | 1.93 | 2.03 | 2.03 |
| 6 | Penicillin | ... | Phenylhydrazine HCl. | ... | 1.15** | 1.45 | 1.53** | 1.93 |
| 7 | Penicillin | ... | Vitamin B ₁₂ and Phenylhydrazine HCl. | ... | 1.12** | 1.66 | 1.91 | 2.16 |
| 8 | Penicillin | ... | Vitamin B ₁₂ | ... | 2.15 | 2.37 | 2.29 | 2.21* |

** $P < .01$ compared to control groups 1 and 5.* $P < .05$ compared to control groups 1 and 5.

TABLE 4

Effect of Vitamin B₁₂ and Antibiotics on Body Temperature, Specific Gravity and Hematocrit of chick blood.

| Serial no. | Treatment | | | | Body temperature °F | | Specific Gravity | | Hematocrit% | |
|------------|-----------------------------|-----|--|-----|---|-------|------------------|-------|-------------|--------|
| | Supplement to basal diet | | Injection | | Time after injection of phenylhydrazine, days | | | | | |
| | | | | | 1 | 7 | 1 | 7 | 1 | 7 |
| 1 | None | ... | None | ... | 106.0 | 106.1 | 1.071 | 1.072 | 21.0 | 22.0 |
| 2 | None | ... | Phenylhydrazine HCl | ... | 106.5 | 106.7 | 1.075 | 1.072 | 12.3** | 21.0 |
| 3 | None | ... | Vita nin B ₁₂ and Phenylhy- drazine HCl. | ... | 106.3 | 106.6 | 1.070 | 1.070 | 16.6** | 24.4 |
| 4 | None | ... | Vitamin B ₁₂ | ... | 105.8 | 106.2 | 1.070 | 1.070 | 24.6 | 26.8** |
| 5 | Penicillin | ... | None | ... | 105.8 | 106.4 | 1.070 | 1.071 | 21.0 | 22.4 |
| 6 | Penicillin | ... | Phenylhydrazine HCl | ... | 105.6 | 106.8 | 1.069 | 1.070 | 14.6** | 24.0 |
| 7 | Penicillin | ... | Vitamin B ₁₂ and Phenylhy- drazine HCl. | ... | 106.2 | 106.7 | 1.070 | 1.071 | 14.6** | 24.6 |
| 8 | Penicillin | ... | Vitamin B ₁₂ | ... | 106.2 | 106.4 | 1.070 | 1.070 | 20.0 | 28.0** |

** $P < .01$ compared to control groups 1 and 5

tions were given. The hematopoietic recovery was faster for those chicks which receive injections of vitamin B₁₂. Chicks receiving injections of both phenylhydrazine HCl and vitamin B₁₂ regained normal hematological levels in 5 days while those receiving phenylhydrazine HCl but not vitamin B₁₂ required 7 days.

Hemoglobin, Erythrocyte counts and Hematocrit values were slightly higher for those chicks that received penicillin. This could be explained on the basis of better growth. No differences were noticed in body temperature and specific gravity of whole blood in any of the groups. Deficiency of vitamin B₁₂ produced slightly lower blood content of hemoglobin, erythrocyte and hematocrit. This shows that this vitamin is needed to maintain normal hemoglobin erythrocyte and hematocrit levels in the blood. This finding is in agreement with that of Swenson (9) who reported that in female chicks, hemoglobin differences significant at 5% level were found between a vitamin B₁₂ deficient group and a second group receiving 3% liver meal. He however, did not get any differences with respect to erythrocyte or hematocrit in male or female chicks. The observations reported in the present paper contradicts the report of Stern *et al.* (6) which reported that the chick was able to maintain normal hemoglobin and hematocrit levels in an uncomplicated vitamin B₁₂ deficiency.

The results on growth are presented in Table 5. Deficiency of vitamin B₁₂ caused a significant retardation of growth which is in general agreement with

TABLE 5
Effect of Vitamin B₁₂ and Antibiotics on weight of chicks at various stages of growth.

| Serial no. | Treatment | | | | Average weight by weeks, gm. | | | |
|------------|--------------------------|-----|---|-----|------------------------------|----|-----|--------|
| | Supplement to basal diet | | Injection | | 0 | 2 | 4 | 6 |
| 1 | None | ... | None | ... | 35.3 | 62 | 160 | 260 |
| 2 | None | ... | Phenylhydrazine HCl | ... | | 60 | 164 | 262 |
| 3 | None | ... | Vitamin B ₁₂ and Phenylhydrazine HCl | ... | | 77 | 192 | 320** |
| 4 | None | ... | Vitamin B ₁₂ | ... | | 76 | 194 | 322** |
| 5 | Penicillin | --- | None | ... | 34.6 | 72 | 192 | 308 |
| 6 | Penicillin | --- | Phenylhydrazine HCl | ... | | 74 | 194 | 310 |
| 7 | Penicillin | ... | Vitamin B ₁₂ and Phenylhydrazine HCl | ... | | 84 | 218 | 364**† |
| 8 | Penicillin | --- | Vitamin B ₁₂ | --- | | 86 | 224 | 368**† |

** P < .01 compared to appropriate basal.

† P < .01 compared to group 3 and 4.

findings of many authors including Stern *et al.* (6). Injection of vitamin B₁₂ to chicks receiving penicillin produced an additional growth response. There was no effect of phenylhydrazine HCl injection on body weight, both in presence and absence of vitamin B₁₂.

SUMMARY

The effect of vitamin B₁₂ and penicillin on hematopoiesis in the chick was studied. Deficiency of vitamin B₁₂ resulted in retardation of growth as well as in a lower blood content of hemoglobin, erythrocyte count and hematocrit. When Vitamin B₁₂ deficiency was aggravated by an anaemia resulting from injection of phenylhydrazine HCl, vitamin B₁₂ was shown to have a stimulatory action on hematopoiesis. A single injection of phenylhydrazine HCl into 6 week old chicks caused anaemia and a seven day period elapsed before hemoglobin, erythrocyte and hematocrit values of deficient chicks reached a normal level whereas only five days were required for hematopoiesis of vitamin B₁₂ supplemented chicks to return to normal.

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SOME PATHOLOGICAL STUDIES OF *PESTALOTIA* SP. CAUSING LEAF SPOT DISEASE OF *LIVISTONA ROTUNDIFOLIA*

By

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Several leaf spot diseases caused by the species of *Pestalotia* have been reported from different parts of the world. Others (1928) described a leaf spot disease of apples caused by *Pestalotia breviseta*. *Pestalotia leprogena* reported by Wardlaw (1934) from Trinidad caused a leaf spot disease of banana.

In India, Mundkur and Keshwala (1942) have reported *Pestalotia citri* from the leaves of *Citrus decummanae*. Chowdhury (1947) found that *Pestalotia palmarum*, caused leaf spot disease of *Borassus flabellifer*. Tandon, Singh and Grewal (1952) studied a leaf spot diseases of *Nephelium litchi* (*Litchi sinensis*) caused by *Pestalotia pauciseta* and soon after Tandon *et al* (1955) studied a leaf spot disease of mango caused by *Pestalotia mangiferae*. Verma (1957) studied the leaf spot disease of *Mimusops elangii* caused by *Pestalotia* sp.

Pestalotia sp. responsible for causing leaf spot disease of *Livistona rotundifolia* is common at Allahabad throughout the year. The plant loses its aesthetic value. The physiological and pathological studies of the organism were undertaken. The present paper deals with the pathological studies only.

MATERIAL AND METHODS

Leaves of *Livistona rotundifolia* (both young and old) were inoculated with *Pestalotia* sp. Pathogenicity tests were conducted on injured as well as on uninjured surface. For isolations the leaf surface was sterilized with 90% alcohol and then with 0.1% mercuric chloride. The leaf surface was then thoroughly washed with sterilized water. Injury was made with sterilized needle. Whenever any infection was observed reisolation was made to confirm the results. The method suggested by Fosberg (1949) was followed for laboratory evaluation of fungicides. The following different methods were used for producing artificial infection on the host.

1. Mass inoculation method.

- (a) Small pieces of inoculum were placed on the injured or uninjured surfaces of leaves.
- (b) The injured or uninjured inoculated area was covered with a moist cotton pad.

2. Spore suspension method.

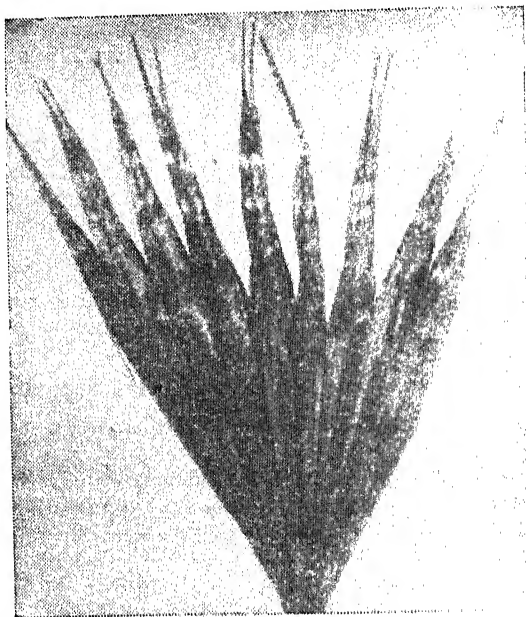
- (a) A suspension of spores in sterilized water was sprayed with an atomizer on the injured or uninjured surfaces of leaves.
- (b) Moist cotton pads were used after spraying.

Cross inoculation were carried out on different hosts. Control measures were tried in the laboratory as well as in the garden. The parts of plant were dusted or sprayed with different fungicides both before and after inoculation. Ridgway's "Color Standards and Color-Nomenclature" was used for determination of colours.

OBSERVATIONS

Symptoms on leaves :

Under natural conditions only the leaves of *Livistona rotundifolia* were infected. At an early stage small circular to elongated spots of deep olive buff colour appear on the leaves and gradually they reach the other surface also (Text Fig. 1). The



Text Fig. 1. Leaf of *Livistona rotundifolia* inoculated with *Pestalotia* sp. (10 days after inoculation).

colour subsequently changes to twany olive and finally it becomes brown. The infected and healthy portions of leaf are separated by a chestnut brown ring. The disease progresses slowly. Pseudopycnidia are mostly found on the lower surface of the leaves and are observed after 15 to 20 days. They appear as raised black pin head like dots on the diseased portion. The diseased area ultimately dries and then it falls off.

Artificial Inoculation :

The symptoms of disease first appear after 3 to 5 days of inoculation. The results are summarized in Table 1 which shows the effect of different methods of inoculation on the appearance of the disease.

TABLE 1
Percentage of infection on leaves of *Livistona rotundifolia* inoculated by
Pestalotia sp.

| Type of inoculum | Surface of leaf | Percentage of infection | |
|---|-----------------|-------------------------|---|
| | | Injured leaf | Uninjured leaf |
| <i>I. Mass inoculation.</i> | | | |
| (a) Without the use of moist cotton pad | Upper | 80% | ... |
| | Lower | 80% | ... |
| (b) With the use of moist cotton pad | Upper | 100% | Infection took place only when the inoculum was placed near the tips. Under such conditions the percentage of infection was 33% |
| | Lower | 100% | |
| <i>II. Spore suspension.</i> | | | |
| (a) Without the use of moist cotton pad | Upper | ... | ... |
| | Lower | ... | ... |
| (b) With the use of moist cotton pad | Upper | 33% | ... |
| | Lower | 20% | ... |

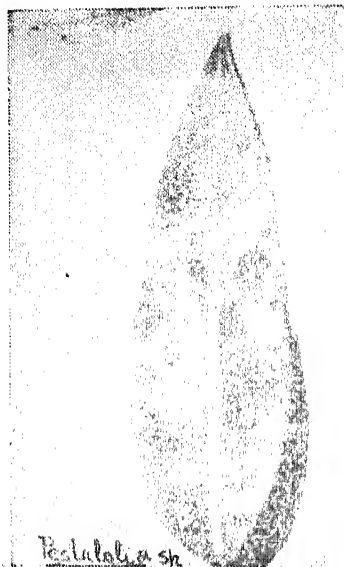
Table 1 indicates that in general the organism did not infect the uninjured leaves except when the inoculum was placed at the tip of the young leaves. The mass inoculation method gave a higher percentage of infection than spore suspension method.

It was further observed that under such conditions the infection was similar on the lower or the upper surface of the leaves. In the absence of moist cotton pads there was no infection by the spore suspension method though the leaves were infected by this method when moist cotton pad was placed over the sprayed region. The percentage infection by this method was very much lower than by the mass inoculation method. Beside the leaves the organism was inoculated on the petiole of *Livistona rotundifolia*. It was observed that the infection was possible at the cut surfaces of the petioles. Isolations from infected portions of leaves and petioles invariably gave *Pestalotia* sp.

Cross inoculation: The organism was inoculated on the leaves of *Pritchardia spinosa*, *Thrinax barbadens*, *Citrus microcarpa*, *Psidium guajava*, *Musa* sp. and *Mangifera indica*. It was observed that inoculation was successful only on the leaves of *Pritchardia spinosa* and *Mangifera indica*. (Text Fig. 2). The fruits of *Psidium guajava* and *Citrus reticulata* were also inoculated but no infection was produced in any of them.

Control method :

Nine fungicides were tested by Fosberg's method in the laboratory. They included Bordeaux mixture 5:5:50, zerlate, phygon, cupravit, copper sandoz, ceresan, tillex, diathane Z-78 and spergon. Out of these zerlate, phygon, ceresan and tillex completely inhibited the growth of *Pestalotia* sp. in the laboratory and they were, therefore, used for garden trials.



Text Fig. 2. Leaf of *Mangifera indica* inoculated with *Pestalotia* sp. isolated from *Livistona rotundifolia*.

The above four fungicides which were found suitable were dusted on the leaves of the host at various intervals (both before and after artificial inoculation). The results are summarized in Table 2.

TABLE 2
Effect of four fungicides dusted on the leaves of *Livistona rotundifolia* inoculated with *Pestalotia* sp.

| Time of inoculation | Fungicides | | | |
|---------------------------------|------------|--------|---------|--------|
| | Zerlate | Phygon | Ceresan | Tillex |
| 1. Just after dusting | - | - | - | - |
| 2. One day after dusting | - | - | + | + |
| 3. Two days after dusting | - | - | + | + |
| 4. Four days after dusting | - | - | + | + |
| 5. Five days after dusting | + | - | + | + |
| 6. Just before dusting | - | - | - | - |
| 7. One day before dusting | - | + | + | - |
| 8. Two days before dusting | + | + | + | + |
| 9. Five days before dusting | + | + | + | + |
| 10. Ten days before dusting | + | + | + | + |
| 11. Fifteen days before dusting | + | + | + | + |

+ or - denotes the appearance or absence of disease respectively,

Table 2 shows that phygon prevented the disease organism to cause the infection even when it was inoculated 5 days after dusting while zerlate prevented infection upto 4 days. It was also evident that none of the fungicides could control the trouble when they were applied 2 days after the organism had become associated with the host. This appears to be due to the migration of the fungus inside the host tissue, where it continues to grow and cause the damage. It was further observed in another control experiment that the disease did not appear whenever phygon was dusted up to a week before the inoculation, but the effect of phygon did not last longer and the disease appeared if this fungicide was applied earlier than a week before the association of the fungus with the host. This clearly shows that if the fungicide is applied long before the organism comes in contact with the host it will not prevent infection.

DISCUSSION

Pestalotia sp. is pathogenic to the leaves of *Livistona rotundifolia*. Guba (1929) stated that species of *Pestalotia* are saprophytic in nature. The present investigations as well as the work of previous investigators like Mundkur and Keshwala (1942) Tandon and Tandon (1948), Tandon, Singh and Grewal (1952) and Tandon *et al* (1955) clearly confirms that the remarks of Guba (l.c.) are not quite correct because a member of species may cause disease.

Mass inoculation with the use of moist cotton pad gave most successful results. Tandon and Bilgrami (1954), Tandon *et al* (1955), Bilgrami (1956) and Verma (1957) also found this method to be most effective for causing the infection on the leaves of various plants studied by them. Mostly the infection could take place only through the injured surface of leaf but infection was also possible from uninjured tips. This indicates that organism could infect young parts of the leaf. Tandon, Sisodia and Bilgrami (1955) had also recorded slight infection on uninjured mango leaves by *Pestalotia mangiferae*. The organism is not very specialized in its parasitic activity as it can cause cross infection in *Pritchardia spinosa* and *Mangifera indica*. Though four of the nine fungicides used in the present investigation could check the growth of the fungus in the laboratory but only one of them (*viz.*, phygon) could control the disease and that too was possible when it was dusted only a week before the organism gets associated with the host. It is often very difficult to estimate the exact period at which the fungus will reach the host. No doubt repeated applications will prevent the leaf spot disease of *Livistona rotundifolia* but the cost will be fairly high. It is, therefore, necessary to search some other fungicide which may remain effective for a longer period.

SUMMARY

Pathogenicity of *Pestalotia* sp. has been established on the leaves of *Livistona rotundifolia*. Cross inoculation were made and it was observed that *Pestalotia* sp. could infect *Pritchardia spinosa* and *Mangifera indica*. Laboratory evaluation of fungicides showed that phygon, zerlate, tillex and cerasan could inhibit the growth of the organism but repeated applications of phygon after every seven days could control the disease.

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SOME PHYSIOLOGICAL STUDIES ON *PESTALOTIA* SP. CAUSING LEAF SPOT DISEASE OF *LIVISTONA ROTUNDIFOLIA*

By

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Diseases caused by *Pestalotia* species have been reported from different parts of the world and they are known to cause leaf spot and fruit rot diseases, but so far detailed physiological investigations have been carried out on a few of them only. Mundkur and Keshwala (1942) have listed 29 species of *Pestalotia* from India and Burma.

Tandon and Bilgrami (1957) working with *Pestalotia mangiferae* found that pH 5.5 to 6.5 was best for the growth and sporulation of their organism. They also found that highly acidic or alkaline medium suppressed sporulation even though the fungus could continue to grow. Verma (1957) observed that most suitable pH for growth and sporulation of *Pestalotia* sp. causing leaf spot disease of *Mimusops elangii* was 5.2.

Shrivastava (1955) found that glucose was best source of carbon for *Pestalotia* sp. causing fruit rot disease of *Citrus meuca var acida*. John (1956) working with two species of *Pestalotia* observed that both species grew best on maltose.

Studies on nitrogen requirements of fungi have shown that various nitrogen compounds have unequal nutritive values for different fungi. John (1956) found that asparagine was the best source of nitrogen for two species of *Pestalotia*.

Verma (1957) reported that ammonium sulphate was best source for *Pestalotia* sp., where as Sisodia (1954) reported that ammonium nitrate was best source for *Pestalotia mangiferae*. Shrivastava (1955) observed that sodium nitrite and potassium nitrite were not toxic to *Pestalotia* sp. He further noticed that peptone was best organic form of nitrogen for his organism.

Phosphorus plays a vital role in the nutrition of fungi. The sources and forms of phosphorus are of importance in assimilation. Tandon (1950) studied the phosphorus requirement of two species of *Pestalotia* and found that organic form of phosphorus (casein and nucleic acid) were most suitable for his organism.

Sisodia (1954) reported that potassium dibasic phosphate was best for the growth and sporulation of *Pestalotia mangiferae*.

Pestalotia sp., responsible for leaf spot disease of palm (*Livistona rotundifolia*) is very common at Allahabad and it causes severe damage. So far it has not been reported on this host. It was, therefore, considered desirable to undertake physiological and pathological studies of the organism responsible for this disease. The results of physiological studies are presented in this paper.

MATERIALS & METHOD

Pestalotia sp. was isolated from diseased leaves of *Livistona rotundifolia*. Single spore cultures were prepared by the dilution method with the help of dummy cutter objective. Asthana and Hawker's medium "A" * was used as a basal medium.

Stock cultures were maintained on the basal medium by subculturing the fungus successively at regular intervals of two weeks. 150 ml conical flasks and guaranteed pure reagents were used throughout the investigation. Liquid cultures containing 25 ml. of nutrient solution in conical flasks were sterilized at 15 lbs pressure for 15 minutes. Fractional sterilization was followed in case of organic media. In every experiment performed on liquid medium, the fungus was allowed to grow for 15 days after which it was harvested on oven dry whatman's filter paper no. 42. After filtration the filter papers containing the mats were kept at 65°C for two days in oven, cooled in desiccator and weighed again. Three replicates of each treatment were taken. Preliminary studies had shown that pH 6.1 was most suitable for the organism, therefore, the pH of the medium was adjusted to the above value in all subsequent experiments. Potassium hydroxide and hydrochloric acid were used in adjusting the pH. Degree of sporulation has been classified on the basis of visual observation (*viz.* excellent, good, fair and poor). Ridgway's "Color Standards and Color Nomenclature" was used for determination of colour. In order to study the effect of different carbon, nitrogen and phosphorus sources, the amount of individual substance in the basal medium was calculated and a quantity equivalent to it was supplied for each substance.

The results were statistically analysed.

OBSERVATIONS

(i) *Effect of different pH concentrations.* The effect of different pH on growth and sporulation was studied. The initial pH of the medium was varied from pH 1.5 to 10.2 (before autoclaving). The results are summarized in Table I.

*Asthana and Hawker's medium A (Basal medium) Glucose 5.0 gms, KNO_3 3.5 gms, KH_2PO_4 1.75, MgSO_4 7H₂O 0.75 gms, Distilled water 1000 CC (2% agar was used for solidification where necessary)

TABLE 1

Showing the dry weight and sporulation of *Pestalotia* sp. at different pH concentrations.

| Different pH | Dry weight in mgms. | Sporulation. |
|--------------|---------------------|--------------|
| 1.5 | 51.0 | Poor |
| 2.1 | 56.0 | Fair |
| 3.2 | 60.0 | Good |
| 4.0 | 62.6 | Good |
| 5.3 | 63.6 | Excellent |
| 6.1 | 65.3 | Excellent |
| 7.0 | 48.6 | Good |
| 7.5 | 44.0 | Good |
| 8.1 | 41.0 | Fair |
| 10.2 | 29.0 | Poor |
| GM : 52.16 | | |

Summary of dry weight results and conclusions at 1% level of P are given below :—

| | |
|------------|--------------------|
| Treatments | Highly significant |
| Replicates | Non significant |
| SE | CD at 1% level |
| 0.86 | ±3.45 |

The various pH values can be arranged in the following order of preference.

Good : pH 6.1, pH 5.3, pH 4.0, pH 3.2 and pH 2.1

Moderate : pH 1.5

Poor : pH 7.0, pH 7.5, pH 8.1 and pH 10.2

The sporulation was best between pH 5.3 to 6.1. The sporulation was poor at both extremes of acidic or alkaline side.

(ii) *Effect of Temperature*. The organism was grown at different temperatures from 6°C to 35°C. It was observed that it was capable of growing at all the above temperatures but statistically the growth and sporulation was best at 21°C. There was no significant difference between 21°C and 27°C while the growth was poor at 10°C and 6°C. There was no marked difference in the thickness of the mycelium at any temperature. Slight difference was observed in the size of spores. It was largest at 27°C ($26.0 \times 6.6 \mu$) and smallest at 35°C ($23.5 \times 5.4 \mu$). It was also noticed that the colour of the median cell was bleached at 35°C and it had changed from dark olive buff to margurite yellow.

(iii) *Effect of different carbon sources*. In order to study the effect of different sources of carbon the following substances were added individually in the basal medium so as to supply 2000 mgms of carbon per litre of nutrient solution.

Arabinose, xylose, glucose, mannose, sucrose, maltose, raffinose, starch, mannitol, sorbitol, tartaric acid and arbutin.

The dry weight and other characters on different carbon sources are recorded in Table 2.

TABLE 2

Showing the dry weight and other characters of *Pestalotia* sp., on different sources of carbon.

| Different carbon sources | Dry weight in mgms | Sporulation | Thickness of Mycelium in μ | Size of spores in μ | Size of Chlamydo spore in μ | Range of Pseudopycnidia in μ |
|--------------------------|--------------------|-------------|--------------------------------|-------------------------|---------------------------------|----------------------------------|
| 1 | 2 | 3 | 4 | 5 | 6 | 7 |
| 1. Arabinose | 23.3 | Poor | 1.9 | 20.3 × 5.3 | — | — |
| 2. Xylose | 62.6 | Fair | 2.2 | 21.9 × 6.3 | — | 100 × 100 |
| 3. Glucose | 71.0 | Excellent | 2.4 | 23.0 × 6.0 | — | 72 × 54 – 162 × 108 |
| 4. Mannose | 72.0 | Good | 2.5 | 23.3 × 6.5 | 5.5 × 6.7 | 54 × 45 – 180 × 162 |
| 5. Sucrose | 80.0 | Good | 2.6 | 22.9 × 6.0 | — | 72 × 90 – 180 × 180 |
| 6. Maltose | 63.3 | Good | 2.4 | 24.5 × 6.1 | — | 90 × 90 – 144 × 162 |
| 7. Raffinose | 74.0 | Good | 2.3 | 22.5 × 6.4 | 5.5 × 5.2 | 37.5 × 45 |
| 8. Starch | 67.0 | Excellent | 2.5 | 23.8 × 6.4 | — | 54 × 54 – 216 × 198 |
| 9. Mannitol | 58.0 | Good | 2.2 | 22.5 × 6.5 | — | — |
| 10. Sorbitol | 74.6 | Good | 2.0 | 22.0 × 6.3 | — | 50 × 50 – 150 × 112.5 |
| 11. Tartaric acid | 62.3 | Fair | 2.1 | 21.8 × 5.8 | — | 45 × 64 – 126 × 108 |
| 12. Arbutin | 59.6 | Fair | 2.3 | 23.4 × 5.9 | — | 54 × 36 – 195 × 142.5 |
| 13. No source carbon | — | — | — | — | — | — |
| GM : 59.79 | | | | | | |

Summary of dry weight results and conclusions at 1% level of P.

| | | |
|------------|-----|--------------------|
| Treatments | ... | Highly significant |
| Replicates | ... | Non significant |
| SE | } | CD at 1% level |
| 2.26 | | ± 8.84 |

In order of preference the above carbon sources may be ground as follows :

Good : Sucrose, sorbitol, raffinose, mannose and glucose.

Moderate : Starch, maltose, xylose, tartaric acid, arbutin and mannitol.

Poor : Arabinose.

The sporulation was best on glucose and starch. It was good on mannose, sucrose, maltose, raffinose, mannitol and sorbitol, fair on xylose, tartaric acid and arbutin and poor on arabinose.

The mycelium was thinnest on arabinose (1.9 μ) and thickest on sucrose (2.6 μ). Though there was no great difference in the size of the spores but they were slightly bigger on carbohydrates except on pentoses. Chlamydo spores were only observed on mannose and raffinose. The pseudopycnidia were not observed on mannitol and arabinose. The largest pseudopycnidia were developed on starch (216 × 198 μ) and smallest on raffinose (37.5 × 45 μ).

The effect of different concentrations of glucose on growth and sporulation was also studied. It was observed that the dry weight continued to increase with

an increase in the amount of glucose from 1 gm. to 50 gms. per litre. There was no growth in complete absence of glucose. The sporulation was best between 10 gms. to 15 gms. of glucose per litre.

(iv) *Effect of different nitrogen sources.* The effect of following nitrogen sources was studied and the substances were added individually in the basal medium, so as to supply 485 mgms. of nitrogen per litre of nutrient solution.

(a) Inorganic-Potassium nitrate, ammonium nitrate, ammonium chloride, ammonium sulphate, magnesium nitrate, sodium nitrate, and sodium nitrite.

(b) Organic : Asparagine, peptone and urea. The dry weight and other characters are recorded in Table 3.

TABLE 3

| Different nitrogen compounds | Dry weight in mgms. | Sporulation | Thickness of mycelium in μ | Size of spores in μ | Size of Chlamydo spores in μ | Range of Pseudopycindia in μ |
|------------------------------|---------------------|-------------|--------------------------------|-------------------------|----------------------------------|----------------------------------|
| 1 | 2 | 3 | 4 | 5 | 6 | 7 |
| 1. Potassium nitrate | 60.3 | Excellent | 2.1 | 24.3 × 6.4 | - | 90 × 108 - 390 × 50 |
| 2. Ammonium nitrate | 39.0 | Excellent | 2.1 | 22.5 × 6.5 | - | 126 × 126 - 252 × 288 |
| 3. Ammonium chloride | 45.6 | - | 2.5 | - | - | - |
| 4. Ammonium sulphate | 55.6 | Excellent | 2.4 | 21.3 × 5.4 | 7.3 × 6.3 | - |
| 5. Magnesium nitrate | 52.0 | Good | 2.0 | 22.8 × 5.6 | - | 162 × 90 - 216 × 160 |
| 6. Sodium nitrate | 46.3 | Fair | 2.1 | 22.5 × 6.3 | - | 144 × 144 - 324 × 184 |
| 7. Sodium nitrate | - | - | - | - | - | - |
| 8. Asparagine | 52.3 | Good | 2.1 | 24.4 × 5.8 | - | 90 × 108 - 648 × 500 |
| 9. Peptone | 45.6 | Poor | 2.2 | 22.5 × 6.3 | - | - |
| 10. Urea | 41.3 | - | 2.4 | - | 7.5 × 10.0 | - |
| 11. No source of nitrogen | Negligible | - | - | - | - | - |
| GM : 43.0 | | | | | | |

Summary of the dry weight results and conclusion at 1% level of P.

| | | |
|------------|-----|--------------------|
| Treatments | ... | Highly significant |
| Replicates | ... | Non significant |
| SE | } | CD at 1% level |
| 2.1 | | ± 8.44 |

In order of preference the above nitrogen compounds may be grouped as follows :

Good : Magnesium nitrate, potassium nitrate, ammonium sulphate and asparagine.

Moderate : Sodium nitrate, ammonium chloride, peptone, urea and ammonium nitrate.

No growth : Sodium nitrate.

The sporulation was best on potassium nitrate, ammonium nitrate and ammonium sulphate. It was good on magnesium nitrate and asparagine, fair on sodium nitrate and poor on peptone. The sporulation was absent on ammonium chloride and urea. There was no marked difference in the thickness of mycelium. The longest spores were observed on asparagine ($24.4 \times 5.8 \mu$). Chlamydospores were observed on ammonium sulphate and urea. Pseudopycnidia were observed only on potassium nitrate, ammonium nitrate, magnesium nitrate, sodium nitrate and asparagine. The size was biggest on asparagine ($648 \times 504 \mu$).

The effect of different concentrations of potassium nitrate was studied and it was found that the growth increase with an increase of potassium nitrate upto 9.0 gms. per litre, but it decreased with any subsequent increase of potassium nitrate. There was significant difference in growth on 9.0 gms. and 15.0 gms. of potassium nitrate per litre. The sporulation was best between 5.0 to 9.0 gms. of potassium nitrate per litre.

(v) *Effect of different phosphorus compounds :*

The following phosphorus compounds were used in order to study the effect on growth and sporulation of *Pestalotia* sp.

Potassium tribasic phosphate, potassium dibasic phosphate, potassium dihydrogen phosphate, sodium phosphate, ammonium phosphate and nucleic acid.

They were added individually in the basal medium so as to supply 399 mgms. of phosphorus per litre of nutrient solution.

The dry weight and other characters are recorded in Table 4.

TABLE 4
Showing the dry weight and other characters of *Pestalotia* sp. on different phosphorus compounds.

| Different phosphorus compounds | Dry weight in mgms. | Sporulation | Thickness of mycelium in μ | Size of spores in μ | Range of pseudopycnidia in μ |
|-----------------------------------|---------------------|-------------|--------------------------------|-------------------------|----------------------------------|
| 1 | 2 | 3 | 4 | 5 | 6 |
| 1. Potassium tri-basic phosphate | 46.6 | Good | 2.6 | 23×6.3 | $36 \times 54 - 90 \times 162$ |
| 2. Potassium dibasic phosphate | 45.1 | Good | 2.8 | 22.3×6.3 | $72 \times 108 - 252 \times 252$ |
| 3. Potassium dihydrogen phosphate | 59.6 | Excellent | 2.2 | 22×6.4 | $54 \times 72 - 180 \times 192$ |
| 4. Sodium phosphate | 43.6 | Good | 2.4 | 23.2×6.3 | $72 \times 72 - 144 \times 180$ |
| 5. Ammonium phosphate | 47.3 | Good | 2.5 | 21.2×6.8 | $54 \times 72 - 144 \times 108$ |
| 6. Nucleic acid | 61.7 | Excellent | 2.2 | 21.3×6.5 | $90 \times 90 - 180 \times 108$ |
| 7. No source of phosphorus | 30.8 | Poor | 2.5 | 20×6.8 | $36 \times 45 - 108 \times 126$ |
| GM : 47.85 | | | | | |

Summary of dry weight results and conclusion at 1% level of P.

| | | |
|------------|-----|--------------------|
| Treatments | ... | Highly significant |
| Replicates | ... | Non significant |
| SE | } | CD at 1% level |
| 1.75 | | ± 7.48 |

The different phosphorus compounds may be grouped in following order of preference.

Good : Nucleic acid and potassium dihydrogen phosphate.

Moderate : Ammonium phosphate, potassium tribasic phosphate, potassium dibasic phosphate and sodium phosphate.

The sporulation was best on nucleic acid and on potassium dihydrogen phosphate. It was poor in complete absence of phosphorus and was good on others. The mycelium was thickest on potassium dibasic phosphate (2.8 μ) and there was no marked difference in others. Similarly there was no marked difference in the size of spores.

Pseudopycnidia were observed in complete absence of phosphorus as well as on all the sources of phosphorus used in the present investigation.

The effect of different concentrations of potassium dihydrogen phosphate was studied. The organism showed best growth on 5.0 gms of potassium dihydrogen phosphate per litre. The growth decreased when the quantity was increased or decreased. The sporulation was best when 1.32 to 1.75 gms potassium dihydrogen phosphate per litre was present.

DISCUSSION

This species of *Pestalotia* was capable of growing within a wide range of pH (viz. 1.5 to 10.2) but it attained best growth and sporulation at pH 6.1. It was also observed that it could grow better on acidic side than on the alkaline side. Similar results were obtained by Shrivastava (1955) and John (1956) working with *Pestalotia* sp. causing fruit rot of *Citrus medica* var *acida* and two species of *Pestalotia* causing leaf spot disease of *Mimusops hexandra* and *Butea frondosa* respectively.

The organism was capable of growing between 6°C to 35°C. Similar results were obtained by Tondon (1947), Sisodia (1954) and John (1956) for *Pestalotia malorum*, *Pestalotia mangiferae* and two species of *Pestalotia*, respectively. 21°C to 27°C was found to be most suitable temperature for growth and sporulation of the fungus and in this respect it was similar to *Pestalotia* sp. isolated from *Camellia japonica* by Ito *et al* (1954). *Pestalotia* sp. could utilize carbon from a number of sources. It was observed that sucrose, sorbitol, raffinose, mannose and glucose were good sources of carbon for this organism.

Verma (1957) working with a species of *Pestalotia* found that sucrose supported best growth of his organism. Sisodia (1954) found that raffinose was best carbon source for *Pestalotia mangiferae* but on the other hand glucose supported poor growth of his organism and in this respect that species differs from the present one. Tandon (1947), Shrivastava (1955) found that glucose supported maximum growth of *Pestalotia malorum*, *P. psidii* and *Pestalotia* species respectively. In the present case glucose is good but is not the best source of carbon.

Maltose which has been found to be a moderate source of carbon for the present organism was found to be the best source for two species of *Pestalotia* studied by John (1956).

Arabinose which has been generally reported to be poor source, was found to be a poor source for the growth and sporulation of this organism also. In this respect the result differed from those of Tandon (1947) who reported that arabinose was a good source for *Pestalotia malorum* and *Pestalotia psidii*.

An increase in the amount of carbon with glucose as a carbon source, increased the growth but it suppressed sporulation. Similar results have been obtained by a number of workers including Verma (1957) who studied *Pestalotia* sp. causing leaf spot disease of *Minusops elangii*.

Asparagine which has been reported to be a good source of nitrogen for two species of *Pestalotia* by John (1956) as well as *Pestalotia* sp. causing fruit rot of *Citrus medica var acida* by Shrivastava (1955) was found to be good source for the present organism. Ammonium sulphate and potassium nitrate which were found to be good sources of nitrogen, have also been reported to be good sources by Verma (1957) for *Pestalotia* sp.

There was no growth on sodium nitrite and in this respect the result differed from that of Shrivastava (1955).

The organism developed best sporulation on ammonium nitrate but failed to sporulate on ammonium chloride. This is interesting specially because pH of both the media was similar.

The present investigations revealed that different carbon and nitrogen sources markedly influenced the sporulation of *Pestalotia* sp. in culture. It ranged from poor to excellent. There was no correlation between growth and sporulation. Pergus (1952), Hacskaylo (1953), Agarwal (1955) and Bilgrami (1956) have also made similar observations.

Amongst the phosphorus sources potassium dihydrogen phosphate and nucleic acid supported good growth of *Pestalotia* sp. Tandon (1950) and Verma (1957) have also found that potassium dihydrogen phosphate and nucleic acid were good sources for *Pestalotia malorum* and *Pestalotia* sp. The sporulation was also best on the above two phosphorus compounds. Shrivastava (1956) also found that potassium dihydrogen phosphate supported good growth and best sporulation for *Pestalotia* sp. studied by him.

The present fungus could grow and sporulate even in absence of any source phosphorus. The result is thus similar to that of Verma (1957) but it differs from that of Sisodia (1954) and Bilgrami (1954) who observed that *Pestalotia mangiferae* and *Phyllosticta cycadina* were incapable of growing in absence of any source of phosphorus.

SUMMARY

An attempt was made to study the physiology of *Pestalotia* sp. causing leaf spot disease of *Livistona rotundifolia*.

The organism could grow and sporulate on a wide range of pH (viz. 1.5 to 10.2) but 6.1 was found to be the most suitable.

Best growth and sporulation of the organism was recorded between 21°C to 27°C.

The fungus was unable to grow in complete absence of carbon. Maximum growth of *Pestalotia* sp. was recorded when 50 gms of glucose per litre was supplied. Sporulation decreased at higher concentrations of glucose. Sucrose was found to be the best source of carbon.

Growth of *Pestalotia* sp. increased with an increase in the quantity of potassium nitrate upto 9.0 gms per litre but any further increase caused decreased growth. Sporulation was best between 5.0 to 9.0 gms of potassium nitrate per litre. Magnesium nitrate was best nitrogen source for the growth of this organism. Potassium nitrate and asparagine were best sources for sporulation of the fungus.

Nucleic acid was the best source of phosphorus for the growth and sporulation of this organism. Sporulation was equally good on potassium dihydrogen phosphate though the growth was inferior. Growth of *Pestalotia* sp. increased even when the concentration of potassium dihydrogen was increased upto 5.0 gms per litre.

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SELECTING GUAVAS FOR WILT RESISTANCE

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The wilt of guava is one of the worst diseases, killing nearly a fourth of guava trees in the orchards of Uttar Pradesh. Since the first appearance of this disease in 1935 in the Babakkarpur area of Allahabad, there is hardly any guava orchard in this State where it has not caused some damage. Its wide spread has been a genuine cause of panic, hence a number of research workers have tried to investigate the cause.

Much of the early work on this disease was confined to isolating micro-organisms from the affected tissues and testing their pathogenicity. Prasad *et al* (1952) finally established that strains of *Cephalosporium* and *Fusarium* caused the disease and that many such strains belonged to the same fungus which they named as *Fusarium oxysporum f. psidii* according to the concept of species in the genus *Fusarium* proposed by Snyder and Hansen (1940). For a satisfactory control of guava wilt, Mathur (1956) emphasised the need for evolving such resistant stocks which may also produce fruits of fine quality. This paper embodies the results of a series of experiments conducted in this direction since 1953 at Kanpur.

It is commonly observed that wilting is most serious in fruit bearing trees and newly planted seedlings and grafts do not show up typical disease symptoms. If natural incidence of disease is observed for a number of years in different varieties, the data thus obtained are usually conflicting, on account of the vagaries of weather from year to year. Trees present a greater difficulty because establishing artificial infection is not easy. Two types of experiments were, therefore, carried out. The first to study the mode of infection and the second to test guava varieties obtained from different sources for resistance to wilt by a technique producing artificial wilting within the shortest possible time.

MATERIALS AND METHODS

Four strains of the fungus *Fusarium oxysporum f. psidii* were used for experiments on the mode of infection and in resistance trials with 47 Indian and foreign guava stocks. Isolate A was taken from a seriously wilted guava tree from an orchard in Babakkarpur area of Allahabad where the disease was first observed in 1935. Isolate C was from a wilted guava tree in the Bithoor area near Kanpur which had produced abundant macro-conidia and was identified and supplied by Dr. N. Prasad, now Plant Pathologist to the Government of Rajasthan. Isolates K and L were from wilted guava trees in the compounds of the Section of the Plant Pathologist to Government of Uttar Pradesh, at Kanpur and Mr. Ali Zaheer now Minister of Finance, Uttar Pradesh, Lucknow, respectively.

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The inoculum was prepared by growing the fungus on maize meal sand medium in 250 c. c. Erlenmeyer flasks for 3 weeks. Liquid cultures of the fungus were prepared in 2 percent potato dextrose liquid medium from which 3 week old spore suspensions were used for injecting into the woody tissues of the guava stems by Roach's method (1934) which was modified for the purpose. Nearly threequarter centimeter wide holes were drilled into the woody tissues of stems of test plants about 2 ft. or more above the ground level. The holes were sterilized with 1 percent mercuric chloride solution before introducing one end of a glass tube, bent at right angles and connected by rubber tubing with a funnel containing the spore suspension. The mouth of the funnel was protected by a petri dish in order to prevent contaminations. About 250 c. c. of the spore suspension was usually sucked in within a fortnight.

The debris of wilted guava trees was prepared by collecting infected plant material including roots, stem and twigs and chopping the infected wood into small pieces 3-5" long and $\frac{1}{4}$ to $\frac{1}{2}$ " thick and burying then about 6" deep into furrows in which guava seedlings were planted. At the time of transplanting a guava seedling in a pit, additional quantities of infected debris were added around the seedlings.

A few week old guava seedlings and grafts used for tests were obtained from the Government Gardens, Allahabad. These seedlings and grafts were raised from guava stocks which bore the finest quality *Safeda*, *Chittidar*, Red flesh and apple shaped guavas. Other Indian guavas used in the tests were Smooth white (Andhra), Smooth green, Red flesh, Lucknow 49, Allahabad and Banarsi obtained from Fruit Research Station, Ananthrajupet, P. O. Cuddapaeh, district of Andhra Pradesh and Dholka, Dharwar, Bombay, Kotharud Sindh and Nasik from Ganesh Khind Fruit Experiment Station, Kirkee, Poona, Bombay State. Dr. W. B. Hayes, Horticulturist, Allahabad Agricultural Institute, supplied seeds of some American varieties acclimated at Allahabad. These were: Stone acid, Popeno, Riverside, Acid, Acid x stone acid, Hart (Naini) and Rolfs (Naini) and their seedlings were raised at Kanpur. Dr. R. A. Hamilton, Assistant Horticulturist, University of Hawaii, College of Agriculture, Honolulu 14, Hawaii, kindly supplied seeds of Fan Retief, Selection P-2, white Guava 6226 and Rolfs (Hawaii), Dr. James W. Lesley, Professor of Genetics, Citrus Experiment Station, Department of Horticulture, Riverside, California, U. S. A., kindly supplied the seeds of Arrows, Detwiller, Horne, Herradura, Hawaiian Sd/9, Webber, Hart (California), Ehrhorn and P.I. 167332. Also seeds of Ruby, Eloina, Supreme, Red Land, Red land Beauty, Fuchs, Clon 32-7, Clon 32-12, Clon 32-18 and Red Indian were kindly sent by Dr. G. D. Ruehle, Vice Director in Charge, Sub tropical Experiment Station, Homestead, Florida, U. S. A. The Horticultural Officer, Peradeniya, Ceylon kindly supplied seeds of China, Safeda and Rolfs California. Thus seeds of Rolfs came from guava plants raised in Hawaii California and Naini (Allahabad) and of Hart from plants grown in California and Naini (Allahabad).

Attempts were made invariably to isolate the pathogen from the inoculated plants.

EXPERIMENTAL WORK

A. Mode of Infection

(a) Placing fungus culture near the root zone :

A few month old seedlings as well as a year and a half old grafts of the *Safeda*, *Chittidar* and apple shaped guavas obtained from Allahabad were planted in 2½ ft. deep pits filled with three fourth soil and one fourth well rotten farm yard manure. A few two year old grafts of seed less guava were also planted. Of these, three plants were selected for tests which were carried out with isolate C of *Fusarium oxysporum* f. *psidii* supplied by Dr. N. Prasad. Three plants of each type were selected for tests.

The fungus was multiplied on maize meal sand medium in 250 c. c. Erlenmeyer flasks. When the culture was 3 weeks old, the contents of the flasks were removed and 250 gm. inoculum was placed near the roots of test plants after removing superficial soil. The roots were then covered with soil which was moistened with water. In this manner the inoculum was placed near the root zone every month beginning from August 1953 to September 1955. Until about six months after inoculation, none of the infected plants showed signs of wilting.

Injecting Spore suspension into Stem wood:

About 6 months old guava seedlings raised from 42 different seed collections obtained from different States of India, United States of America, Hawaii and Ceylon were planted on September 29, 1954 and when they were about a year and half old, their stems were injected about 18" from the ground level, with 100 c.c. spore suspension prepared from a 3 week old culture of isolate C of *Fusarium oxysporum* f. *psidii*.

This method of inoculation also failed because no mortality was observed in the injected plants until June 30, 1956.

Infection by debris from wilted plants :

Naturally wilted plants were up rooted and their roots, stems and twigs were chopped into small pieces and buried 6" deep near the root zone of the test plants. The fine roots were partly injured. In one experiment of this type thirteen, 5 year old *Safeda* plants were treated with naturally infected debris in the middle of August 1953.

Wilting in treated trees was first observed in 4 plants by the first week of October 1954 i.e. after about one year and two months. A year later all the plants died successively between May and October 1955.

This mode of infection being the most satisfactory, a small wilt sick plot was prepared by burying infective debris into it on July 30, 1955. More debris was later on added to the wilt sick plot on December 18, 1956, April 20, 1957 and frequently during 1958 and 1959. Test plants were grown in this plot and the mortality of plants was recorded regularly. The results are given under experiments on Varietal Resistance Trials.

B. Varietal Resistance Trials

While studies on the mode of infection were in progress, varietal trials were also conducted in pots, wilt sick plots and in the field using different methods to create maximum infection in the varieties in order to test their resistance to the wilt disease caused by *Fusarium oxysporum* f. *psidii*.

In September, 1954, 7-10 month old seedlings belonging to 44 varieties were transferred to pots. Each pot had one seedling and 4 seedlings of each variety were inoculated with isolates A, C, K, and L of the causative fungus. One seedling of a variety was inoculated with one strain of the fungus. The technique of inoculation was to place every month for about a year, 250 gms. of 3 week old cultures multiplied on maize meal sand medium. The plants that showed initial wilting are marked with (+) and those remained healthy until a few months after the last inoculation are

marked (■) in Section A of Table 1. By the end of 1955, these plants were transplanted in the field where some recovered from initial wilting and others died due to virulence of the pathogen within the plant. The latest mortality and survival of plants in December 1959 is given in Section B of Table 1. It is also clear from this table that isolate C is not parasitic whereas isolates A, K and L are highly parasitic.

Also in September 1954, three seedlings, each 6 months old belonging to 42 varieties were planted in a nursery and a year later i.e. in September 1955 when they were 1½ years old they were injected into the stem with spore suspensions of the causative fungus. But since no mortality was observed until June 30, 1956, they were transplanted in an orchard and further treated with debris of infected material at the base. About the same time 5 plants, each of 17 varieties were also planted in an adjoining orchard and treated with debris of naturally wilted guava trees. In due course some plants began to die and a progressive total of the mortality observed in December 1959 is given in Table 2. In the same table are recorded the results of wilt resistance of 47 varieties in a wilt sick plot. In table 3, a list of guava varieties most tolerant to wilt together with their growth characters has been given. The fruiting characters of these wilt tolerant selections remain to be studied by the Horticulturist, but in the meantime a few grafts of the famous Allahabad *Ghittidar* and *Safeda* guavas have been established on some of them.

Further work is necessary to produce a sufficiently large number of wilt resistant grafts which could bear fruits of fine quality in the orchards of fruit growers.

TABLE 1
Infectivity Tests of Indian and Foreign Guavas with four Isolates of
Fusarium Oxysporum F. *Psidii*

| Sl. No. | Name and Source of Variety | Section A | | | | Section B | | | |
|---------|--------------------------------|---|------------|---|---|----------------|---|---|---|
| | | Nursery, 1954-56 | | | | Field, 1956-59 | | | |
| | | Fungus isolates | | | | | | | |
| | | A | C | K | L | A | C | K | L |
| 1. | Fan Retief (Hawaii) | - | + | + | + | + | | | + |
| 2. | Selection P. 2(„) | + | - | - | + | + | + | + | |
| 3. | No. 6229 white („) | + | - | - | - | | - | + | + |
| 4. | Rolfs („) | + | - | + | - | + | + | + | + |
| 5. | Arrons (California) | + | - | - | + | + | | - | - |
| 6. | Detwiler („) | - | - | - | - | + | - | | + |
| 7. | Horne („) | No germination, hence no tests conducted. | | | | | | | |
| 8. | Herradura („) | + | Not tested | | + | + | - | - | + |
| 9. | Hawaii an Sd/9 („) | + | - | - | + | + | - | + | - |
| 10. | Webber („) | + | - | - | + | | + | - | |
| 11. | Hart („) | + | - | - | + | | + | + | |
| 12. | Ehrhorn („) | + | - | - | + | - | - | + | |
| 13. | P. I. 167332 („) | No tests conducted | | | | | | - | + |
| 14. | Ruby (Florida) | + | - | - | + | + | - | + | - |
| 15. | Eloina („) | - | - | - | - | + | + | + | - |
| 16. | Supreme („) | - | - | - | + | + | - | + | + |
| 17. | Red land („) | + | - | - | + | | - | - | - |
| 18. | Red land Beauty (Florida) | + | - | - | + | + | - | - | |
| 19. | Fuchs („) | + | - | - | + | | - | - | - |
| 20. | Clon 32-7 („) | - | - | - | - | + | - | + | + |
| 21. | Clon 32-12 („) | - | - | - | - | - | - | + | + |
| 22. | Clon 32-18 („) | - | - | - | + | - | + | - | + |
| 23. | Red Indian („) | + | - | - | - | + | - | + | + |
| 24. | Stone Acid („) | + | - | - | + | - | - | + | + |

(+) indicates mortality and (-) indicates a healthy plant.

TABLE 1 (contd.)
Infectivity Tests of Indian and Foreign Guavas with four Isolates of
Fusarium Oxysporum F. Psidii

| S. No. | Name and Source of Variety | Section A | | | | Section B | | | |
|--------|------------------------------------|------------------|---|---|---|----------------|---|---|---|
| | | Nursery, 1954-56 | | | | Field, 1956-59 | | | |
| | | Fungus isolates | | | | | | | |
| | | A | C | K | L | A | C | K | L |
| 25. | Popeno (Florida) | + | - | - | + | + | - | + | + |
| 26. | Riverside (California) | + | - | - | + | | - | + | + |
| 27. | Acid („) | + | - | - | + | | - | + | + |
| 28. | Acid x Stone/ Acid (California) | - | - | - | - | - | - | - | + |
| 29. | Smooth white(Andhra) | + | - | + | + | | - | + | - |
| 30. | Smooth green(„) | - | - | - | + | + | - | + | |
| 31. | Red fleshed („) | - | - | - | - | + | - | | + |
| 32. | Lucknow-49 („) | - | - | - | + | + | - | + | + |
| 33. | Allahabad („) | + | + | - | + | + | + | - | - |
| 34. | Benarsi („) | + | - | + | + | | - | + | |
| 35. | China (Ceylon) | Not tested | | | | | | | |
| 36. | Safeda („) | + | - | - | - | + | - | + | |
| 37. | Rolis (California) | + | - | - | - | + | - | + | + |
| 38. | Webber (Florida) | + | - | + | - | + | + | + | + |
| 39. | Hart (Naini) | + | - | - | + | - | - | + | |
| 40. | Rolfs („) | - | - | - | - | + | + | | + |
| 41. | Dholka (Bombay) | - | - | + | - | - | + | + | + |
| 42. | Dharwar („) | - | - | + | - | - | + | | - |
| 43. | Kotharud („) | - | - | + | - | + | + | | - |
| 44. | Sindh („) | + | - | + | + | + | + | | + |
| 45. | Nasik („) | + | - | + | - | | | | |
| 46. | Safeda (Allahabad) | + | + | + | + | | | | |
| 47. | Red flesh („) | - | - | - | - | | | | |
| 48. | Chittidar („) | Not tested | | | | | | | |
| 49. | Seedless grafts(„) | Not tested | | | | | | | |

(+) indicates mortality and (-) indicates a healthy plant.

TABLE 2
Field resistance of Indian and Foreign Guava Clones to wilt caused by *Fusarium Oxysporum F. Pasidi*

| Sl. No. | Name and Source of variety | In field from 1956-1959 | | | | Total No. of seedlings and grafts planted | No. of seedlings and grafts wilted | No. of seedlings and grafts surviving |
|---------|----------------------------|--|-----------------------------|----------------|----------------|---|------------------------------------|---------------------------------------|
| | | 1 | 2 | 3 | Debris only | | | |
| | | Wilt-nursery | Spore-suspension and Debris | Debris | | | | |
| | | Wilted/Healy | Wilted/Healthy | Wilted/Healthy | Wilted/Healthy | | | |
| 1 | Fan Retief (Hawaii) | 1/3 | 2/3 | ... | | 6 | 3 | 3 |
| 2 | Selection P-2 (") | 0/3 | 2/3 | 3/5 | | 10 | 5 | 5 |
| 3 | No. 6229 white guava ") | 1/1 | 0/3 | ... | | 4 | 1 | 3 |
| 4 | Rolfs (") | 2/4 | 1/3 | 3/5 | | 12 | 6 | 6 |
| 5 | Arrons (California) | 3/4 | 1/3 | 1/5 | | 12 | 5 | 7 |
| 6 | Detwiler (") | 1/4 | 1/3 | 3/5 | | 12 | 5 | 7 |
| 7 | Horne (") | No germination, hence no tests conducted | | | | | | |
| 8 | Herradura (") | 1/4 | 1/3 | ... | | 7 | 2 | 5 |
| 9 | Hawaiian Sd/9 (") | 0/4 | 2/3 | 4/5 | | 12 | 6 | 6 |
| 10 | Webber (") | 2/4 | 2/3 | 1/5 | | 12 | 5 | 7 |
| 11 | Hart (") | 2/3 | 2/3 | 3/5 | | 11 | 7 | 4 |
| 12 | Ehrhorn (") | 2/4 | 1/3 | 3/5 | | 12 | 6 | 6 |
| 13 | P. I. 167332 (") | ... | 1/3 | ... | | 3 | 1 | 2 |
| 14 | Ruby (Florida) | 2/4 | 2/3 | 1/5 | | 12 | 5 | 7 |
| 15 | Eloina (") | 2/4 | 0/3 | 2/5 | | 12 | 4 | 8 |
| 16 | Supreme (") | 1/4 | 0/3 | 1/5 | | 12 | 2 | 10 |
| 17 | Red land (") | ... | 2/3 | 4/5 | | 8 | 6 | 2 |
| 18 | Red land Beauty (") | 2/4 | 1/3 | 4/5 | | 12 | 7 | 5 |
| 19 | Fuchs (") | 0/4 | 1/3 | 3/5 | | 12 | 4 | 8 |
| 20 | Clon 32-7 (") | 4/4 | 0/3 | 2/5 | | 12 | 6 | 6 |

| | | | | | | | | |
|----|-------------------|--------------|------------|-----|-----|----|---|---|
| 21 | Clon 32-12 | (") | ... | 0/3 | 2/5 | 8 | 2 | 6 |
| 22 | Clon 32-18 | (") | 1/4 | 1/3 | ... | 7 | 2 | 5 |
| 23 | Red Indian | (") | 2/4 | 3/3 | ... | 7 | 5 | 2 |
| 24 | Stone Acid | (") | 0/4 | 3/3 | ... | 7 | 3 | 4 |
| 25 | Popeno | (") | 0/4 | 1/3 | ... | 7 | 1 | 6 |
| 26 | Riverside | (California) | 1/4 | 0/3 | ... | 7 | 1 | 6 |
| 27 | Acid | (") | 0/4 | 3/3 | ... | 7 | 3 | 4 |
| 28 | Acid x Stone Acid | (") | 1/4 | 1/3 | ... | 7 | 2 | 5 |
| 29 | Smooth white | (Andhra) | 0/4 | 2/3 | ... | 7 | 2 | 5 |
| 30 | Smooth green | (") | 3/4 | 0/3 | ... | 7 | 3 | 4 |
| 31 | Red fleshed | (") | 1/4 | 3/3 | ... | 7 | 4 | 3 |
| 32 | Lucknow-49 | (") | 1/4 | 1/3 | ... | 7 | 2 | 5 |
| 33 | Allahabad | (") | 1/4 | 1/3 | ... | 7 | 2 | 5 |
| 34 | Benarsi | (") | 0/4 | 0/3 | ... | 7 | 0 | 7 |
| 35 | China | (Ceylon) | Not tested | | ... | | | |
| 36 | Safeda | (") | 0/4 | 1/3 | ... | 7 | 1 | 6 |
| 37 | Rols | (California) | 0/4 | 0/3 | ... | 7 | 0 | 7 |
| 38 | Webber | (Florida) | 0/4 | 1/3 | ... | 7 | 1 | 6 |
| 39 | Hart | (Naini) | 0/4 | 0/3 | ... | 7 | 0 | 7 |
| 40 | Rols | (Naini) | 0/4 | 1/3 | ... | 7 | 1 | 6 |
| 41 | Dholka | (Bombay) | 0/4 | 0/3 | ... | 7 | 0 | 7 |
| 42 | Dharwar | (") | 2/4 | 0/3 | ... | 7 | 2 | 5 |
| 43 | Kotharud | (") | 0/3 | 1/3 | ... | 6 | 1 | 5 |
| 44 | Sindh | (") | 0/3 | 0/3 | ... | 6 | 0 | 6 |
| 45 | Nasik | (") | 0/3 | 1/2 | ... | 5 | 1 | 4 |
| 46 | Safeda | (Allahabad) | 2/4 | 4/6 | ... | 10 | 6 | 4 |
| 47 | Red flesh | (") | 4/4 | ... | 2/5 | 9 | 6 | 3 |
| 48 | Chittidar | ... | ... | 2/5 | ... | 5 | 2 | 3 |
| 49 | Seedless graft | ... | ... | 2/3 | ... | 3 | 2 | 1 |

TABLE 3

Guava clones most tolerant to wilt caused by *Fusarium Oxysporum* *F. Psidii*

| Serial No. | Name and source of variety | Total No. of seedlings and grafts planted | No. of seedlings and grafts wilted | No. of seedlings and grafts surviving | Remarks |
|------------|--------------------------------|---|------------------------------------|---------------------------------------|---|
| 1 | No. 6229 white guava (Florida) | 4 | 1 | 3 | Shrub 5' - 2" Fruits smaller than the prevalent Safeda variety. |
| 2 | Supreme (Florida) | 12 | 2 | 10 | Tree 11' - 6" Fruits smaller than the Safeda variety. |
| 3 | Clon 32-12 (Florida) | 8 | 2 | 6 | " " " |
| 4 | Popeno (Florida) | 7 | 1 | 6 | Tree 12' - 6" Fruits smaller than Safeda. |
| 5 | Riverside (California) | 7 | 1 | 6 | Tree 14' - 6" Fruits smaller than Safeda. |
| 6 | Benarsi (Andhra) | 7 | 0 | 7 | Tree 12' - 6" Fruits about the size of the Safeda variety. |
| 7 | Safeda (Ceylon) | 7 | 1 | 6 | Tree 9' - 4" Fruits smaller than the Safeda variety. |
| 8 | Rolfs (California) | 7 | 0 | 7 | Tree 10' - 2" Fruits much smaller than the Safeda variety. |
| 9 | Webber (Florida) | 7 | 1 | 6 | Tree 16' - 12" Fruits smaller than the Safeda variety. |
| 10 | Hart (Naini) | 7 | 0 | 7 | Tree 15' - 6" Fruits of almost the size of Safeda variety. |
| 11 | Rolfs (Naini) | 7 | 1 | 6 | Tree 10' - 6" Fruits smaller than the Safeda variety. |
| 12 | Dholka (Bombay) | 7 | 0 | 7 | Tree 13' - 4" Fruits smaller than the Safeda variety. |
| 13 | Sindh (Bombay) | 6 | 0 | 6 | Tree 10' - 6" Fruits smaller than the Safeda variety. |
| 14 | Nasik (Bombay) | 5 | 1 | 4 | Tree 12' - 6" Fruits smaller than the prevalent Safeda variety. |

SUMMARY

Wilt of guava is caused by *Fusarium oxysporum f. psidii*. Experiments were conducted to study different modes of infection viz. soil inoculation with fungus culture, with or without injury to the roots, injection of spore suspension into the stem and addition of debris of small pieces of naturally wilted guava trees near the root zone of healthy plants. The addition of debris was found most satisfactory for causing wilt within a short time.

Varietal trials for wilt resistance in 47 varieties of guava obtained from different sources in India and abroad were conducted in pots, wilt-sick plots and in the field. Tests with different modes of infection show that varieties. White guava, No. 6229, Supreme, Clon 32-12, Webber and Popeno from Florida, U. S. A., Hart and Rolfs from Florida, but acclimated at Allahabad, Riverside and Rolfs from California, U. S. A., *Safeda* from Ceylon and Benarasi (Andhra strain), Dholka, Sindh and Nasik (Bombay) from India are the most tolerant to the disease.

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STUDIES ON INDIAN *ICHNEUMONIDAE* (PARASITIC HYMENOPTERA)

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Description of three new species and new records of eight known species of Indian *Ichneumonidae*, Parasitic Hymenoptera, form the subject matter of this paper.

This paper forms a part of the studies undertaken to work out the specimens of hymenopterous parasites received for identification from various sources. We have included in this paper descriptions of three new species and records of as many as eight known *Ichneumonidae*. We take this opportunity of expressing our grateful thanks to Dr. N. G. Shabde, Principal, College of Science, Nagpur and to Dr. S. M. H. Khatib, Professor of Zoology, also of College of Science, Nagpur for all the facilities provided to us in the course of the present work.

Type specimens and slides are for the time being kept in the collections of the authors.

Subfamily *PIMPLINAE*

Tribe *Pimplini*

Xanthopimpla indica, sp. nov.

♀ Body yellowish-brown, with black markings. Head viewed from above broadly oval, thrice as broad as long, distinctly constricted behind eyes, ocelli brown, large, in a triangle on black background; interocellar space equal to the ocellular, front ocellus as far away as the interocellar space, interorbital space one third the breadth of head, vertex yellow, shiny, convex, smooth, with a black median spot; occiput margined behind, shiny, smooth, black, convex. Head viewed from front (fig. 1) nearly circular, shiny, naked, inner orbital border sinuate just above the insertion of antenna, frons shiny, smooth, without pubescence, yellow except for the black marking, continued from a similar marking on vertex on to the middle of antennal scrobes; antenna (figs. 4, 5 and 6) inserted above face, a little shorter than body, reddish brown, filiform, gradually tapering towards apex, lighter towards apex than at base, with 43 segments, flagellate segments all cylindrical, scape (fig. 4) yellow below on the outer side, black above and on inner side, pedicel (fig. 4) short and cylindrical; first flagellar segment (fig. 4) as long as scape and pedicel combined, black on one side and yellow on the other, flagellate segments 7—10 as in figure 6 and segments 36—43 as in figure 5, terminal segment a little less than twice the length of penultimate segment, sparsely pubescent; face flattened dorsally, moderately covered with short white pubescence, depressed mesially a little below antennal scrobes, with distinct lateral foveae on the lower third of face, yellow, rugosely punctate, clypeus clearly discrete, moderately convex, with a strongly concave apex, sparsely pubescent, yellow; mandible large, yellow with black apices, palpus yellow. Head viewed from side oval, gena short, one-eighth the length of eye, covered with white pubescence, temples broad half the width of eye in the middle.

Thorax yellow with black markings, slightly raised above the level of head, pronotum shiny, sparsely pubescent, smooth, mesonotum shiny, with three large black markings across the anterior region and a fourth posteriorly in middle, laterally strongly margined, scutellum strongly convex, nearly rectangular, strongly

margined, raised a little above the level of mesothorax, smooth, glabrous, postscutellum small, glabrous, convex, yellow, smooth, without pubescence, propodeum with closed hexagonal areola issuing off lateral carinae, with back marking anteriorly on the sides, glabrous, clearly striate, without pubescence, with two moderate, median carinae, spiracle large, linear, obliquely situated near middle. Legs short, stout, finely pubescent, yellow, hind legs longer than the rest, coxa a little longer than broad, longer than both the trochanters combined, trochanter twice as long as trochanterellus, femur stout, longer than coxa and trochanters combined, two and three-fourth times as long as broad, tibia longer than femur, black at base, with two subequal apical spurs (fig. 2), gradually widened at apex, metatarsus nearly one-fourth the tibia, tibial spur five-sevenths the length of metatarsus, claw (fig. 3) simple, stout, dark brown. Wings (figs. 7 & 8) hyaline, thrice as long as broad, veins brown, pterostigma light brown, with a pale patch between pro- and pterostigma, narrow, long, cell R_2 long, narrow, apical abscissa of $SR + R_2$ distinctly curved at apex, cell R_5 small, quadrate, with a short petiole, vein M_2 emitted from middle, curved, R_3 incomplete, rest of details and hind wing as in figures 7 and 8.

Abdomen petiolate, deplanate, two thirds the length of body, finely pubescent, tergites closely shallowly punctate, broader than long, first tergite broader than long, narrow at base, with two lateral large black markings, at the apical three-fourths, middle region raised with two longitudinal ridges, raised area broader at base, narrow apically, spiracle large, broadly linear; second tergite broader than long, shorter than first tergite, shallowly closely punctate, sparsely pubescent, with a slightly raised rhomboidal area in middle, broader apically than at base, spiracle broadly linear; third tergite twice as broad as long, broadest at apex, more definitely punctate than second tergite, sparsely pubescent, with two black markings, faint striae in between the markings, apically with a transverse ridge; fourth tergite similar to third tergite, striae more clear, pubescence and punctation more prominent than in the basal segments, third and succeeding segments with a transverse ridge at apex; fifth tergite with an additional small faint, narrow black marking in middle, anteriorly on either side of this marking are two striate brown areas, pubescence and punctation more prominent than in the basal segments; sixth segment with very faint and small black markings; seventh segment with much bigger black markings, bigger than in any of the preceding segments; eighth segment with two very small markings; terminal segments telescoped: terebra exerted, moderately long.

Length ♀ 15 mm.

Holotype one ♀ on pin labelled "Loc. Forest, Host : Joma, Date : Reared, Coll. St. A. G. N. D." (Material received from the Agricultural College, Bapatla Andhra Pradesh). Antenna, wings and legs mounted on slides.

This species has close superficial resemblance to *hornata* Cameron and *pedator* Fabricius (1). It is different from *hornata* in having two black and very big markings on the seventh tergite and from *pedator* in the apical abscissa of the radial vein not being sinuate but only curved at apex. It also differs from both these species in many details of the antennal and thoracic structures.

Subfamily Ophioninae

Tribe Ophionini

Enicospilus silvaraji Rao and Kurian

1951. *Enicospilus silvaraji*, Rao and Kurian, *Indian J. Ent.*, 13(1): 28-31.

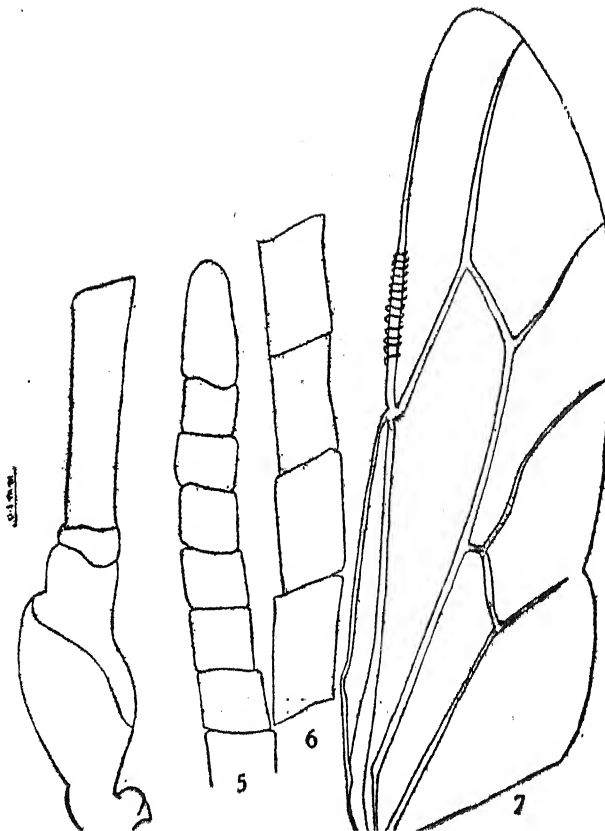
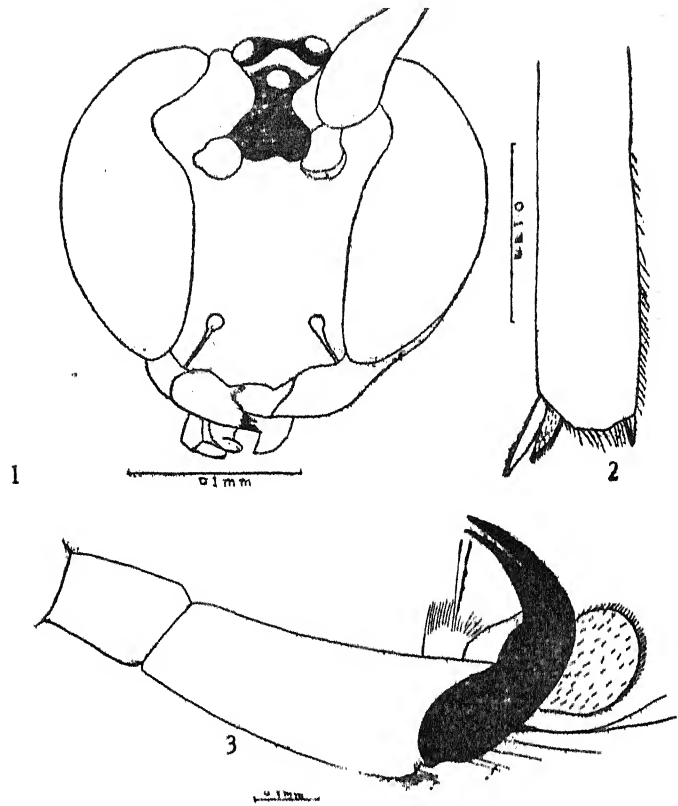


Fig. 1—7. *Xanthopimpla vindica*, sp. nov. 1. Head viewed from front, 2. Hind tibial spurs, 3. Hind claw, 4. Scape, pedicel and first flagellar segment, 5. Terminal antennal segments, 6. Seventh to tenth antennal segments and 7. Hind wing.

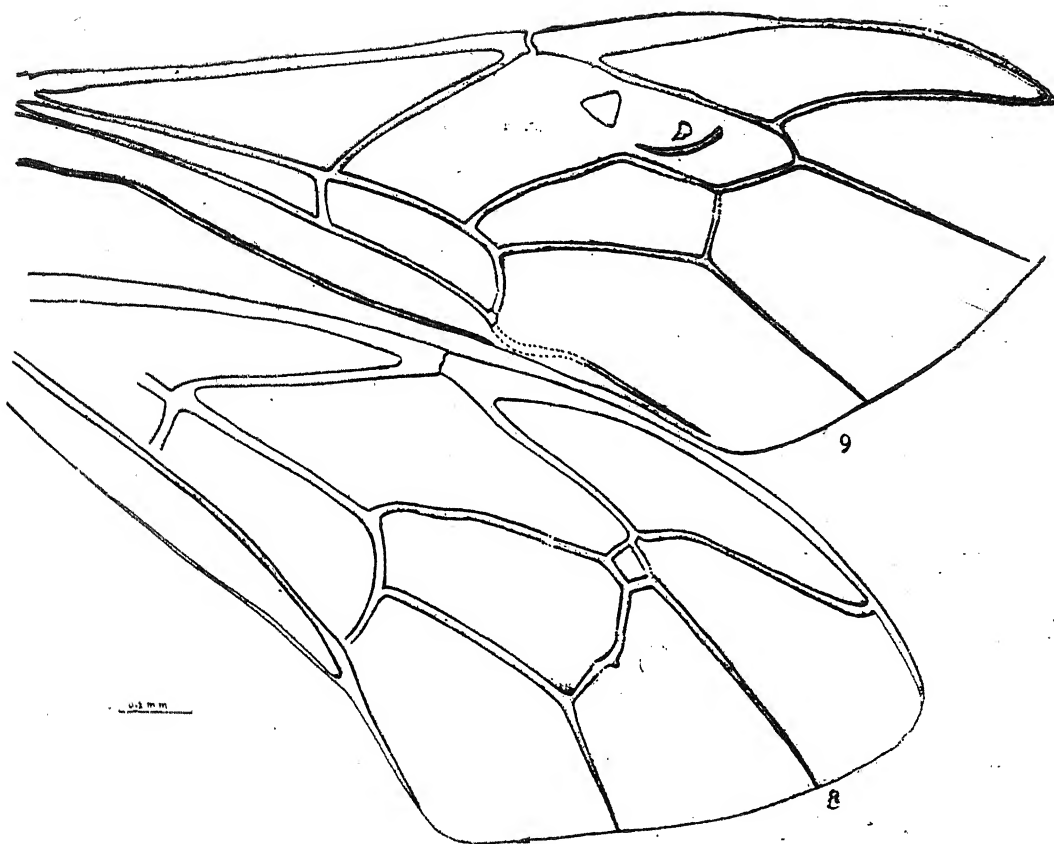


Fig. 8. Fore wing of *Xanthopimpla indica*, sp. nov., and 9. Fore wing of *Enicospilus indica*, sp. nov.

From among the collections under study are found two ♀♀ and one ♂ of this species labelled "SNR Coll., Dining Room, at light, 18-2-1958, Ravinagar, Nagpur." These resemble the described species in all details but appear a little large. ♂ 16 mm. and ♀ 17.5 mm.

This species, previously recorded from Agra (U. P.) is for the first time being reported from Nagpur (Vidarbha), Bombay State.

This species is reported to be parasitic on *Adisura atkinsoni* Moor in Mysore.

Enicospilus rufus Tosquinet

1846. *Ophion rufus*, Brulle, *Hist. Nat. Ins. Hym.*, 4: 149.

1896. *Ophion*-(*Enicospilus*) *rufus*, Tosquinet, *Ichn. d'Afrique*, p. 378 (♂♀).

1913. *Henicospilus rufus*, Morley, *Faun. Brit. India, Hymen.*, 3: 385.

We refer to this species one ♀ labelled "At light, S. G. Hardas, Sept. 1956". This species was previously recorded from Assam, Tenasserim, Ceylon, China, Java, Bourbon and Africa. This is the first time we have come across this species from Nagpur (Vidarbha), Bombay State.

Enicospilus indica, sp. nov.

♀ Body light brown, posterior part of abdomen darker. Head viewed from above (fig. 18) quadrate, yellowish brown, slightly narrower than thorax, thrice as long as broad, ocelli prominent, interocellar space nearly equal to ocellular space and thrice the diameter of ocellus, interorbital space half the width of head, vertex convex, margined, smooth, pubescent, occiput excavate. Head viewed from front (fig. 17) broadly oval, frons yellowish-brown, convex and smooth, face rugosely punctate, pubescent, yellowish-brown in middle, paler on the sides, convex, clypeus moderately convex, not margined, rugosely punctate, brown, sparsely pubescent, lateral foveae shallow, mandible dark reddish-brown, apices black, palpi yellow, pubescent. Head viewed from side oval, temples one-third the width of eye, gene short, one-ninth the height of head. Antenna (figs. 13, 14 and 15) inserted above the middle of face, filiform, reddish-brown, with 46 segments (49 segments in some paratypes), shorter than body, slightly tapering towards apex, flagellate antennal segments cylindrical, scape (fig. 13) cylindrical, sparsely pubescent, longer than pedicel, the latter (fig. 13) broader than long, first flagellate segment (fig. 13) longer than scape and pedicel combined, segments 6-8 and 40-42 as in figures 14 and 15 respectively, terminal segment shorter than penultimate. Eyes sinuate opposite to the insertion of antenna, naked.

Thorax reddish-brown, broader than head, mesonotum closely finely punctate, pubescent, scutellum moderately convex, nearly triangular, moderately pubescent, rugoso-punctate, laterally margined, lighter than mesothorax, mesopleura finely closely punctate, pubescent, mesially distinctly striate, postscutellum thickly margined posteriorly, propodeum rugoso-punctate, finely pubescent, without an areola and carinae, petiolar area distinctly marked, spiracle large, linear. Legs long slender, reddish-brown, covered with short pubescence, coxa (fig. 11) smooth, sparsely pubescent, slightly longer than both the trochanters combined, trochanter (fig. 11) nearly twice the trochanterellus, femur finely pubescent, much longer than coxa and trochanters combined, tibia slender, longer than femur, covered by stiff setae, tibial spurs (fig. 12) unequal, mesially brown, light reddish-brown on sides,

longer spur reddish-brown, one-third the length of metatarsus, foliaceously expanded in the basal one-fourth, shorter spur half the length of longer spur, tarsal segments covered by short stiff setae, metatarsus finely pubescent, shorter than rest of tarsal segments combined, with short stiff spines on the inner side, claw (fig. 10) pectinate. Wings (figs. 9 and 10) large, membranous, except for the three corneous pale yellow marks, two and a half times as long as broad, veins brown, pterostigma narrow, emitting the radius before its middle, shorter than R_1 , vein $SR+R_3$ wanting, vein r swollen and slightly sinuate in middle, slightly curved at the basal one-fourth, cell R_5 with clear space except for the three corneous markings, largest of the three triangular, second slightly smaller, also triangular, longer than broad, third long, linear, curved, vein M_{3+4} conspicuously curved, apical one-fourth unpigmented, M_2 issued before R_5 , vein M_4+Cu_1 slightly behind $m-cu$, rest of the details and hind wing as in figures 9 and 10. Spiracle circular, small situated in the apical third.

Abdomen more than twice the length of head and thorax combined, laterally strongly compressed, petiolate, reddish-brown, except for the darker apex, with silvery white pubescence first tergite long, linear, parallel sided upto the apical third, broader onwards, smooth, shiny, second tergite short, broader than the first, broadest in middle, rest of the tergites gradually becoming shorter and telescoped, ovipositor sheath exerted.

♀ length 13 mm.

Holotype 1 ♀ pinned and labelled "S. N. Rao Coll., at light, 15-2-58, Dinning table, Ravinagar, Nagpur". Wings, legs and antenna mounted on slides.

Paratypes 2 ♀♀ pinned and labelled "Coll. M. T. Damle, 10-10-55, on wing, Dantoli, Nagpur" and "Coll. P. Grover, amidst clothing, Nagpur, Jan. 1959". Wings legs and antenna mounted on slides.

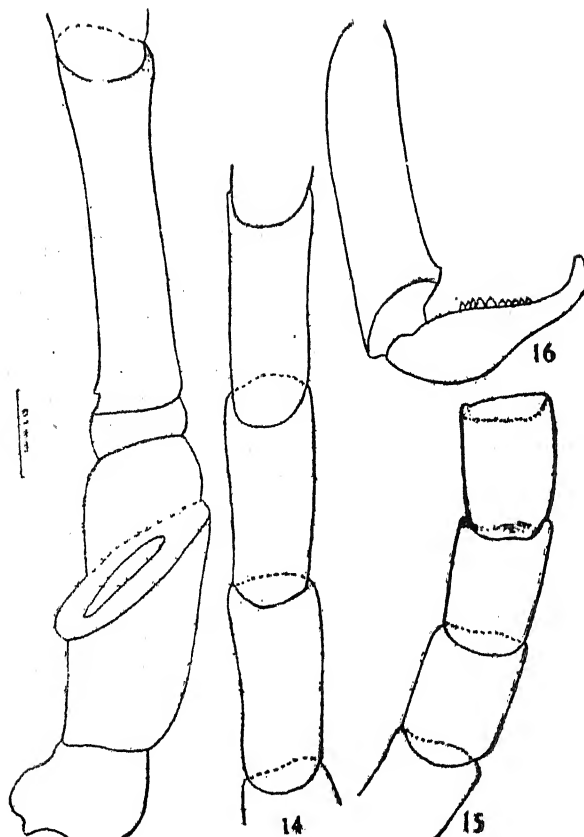
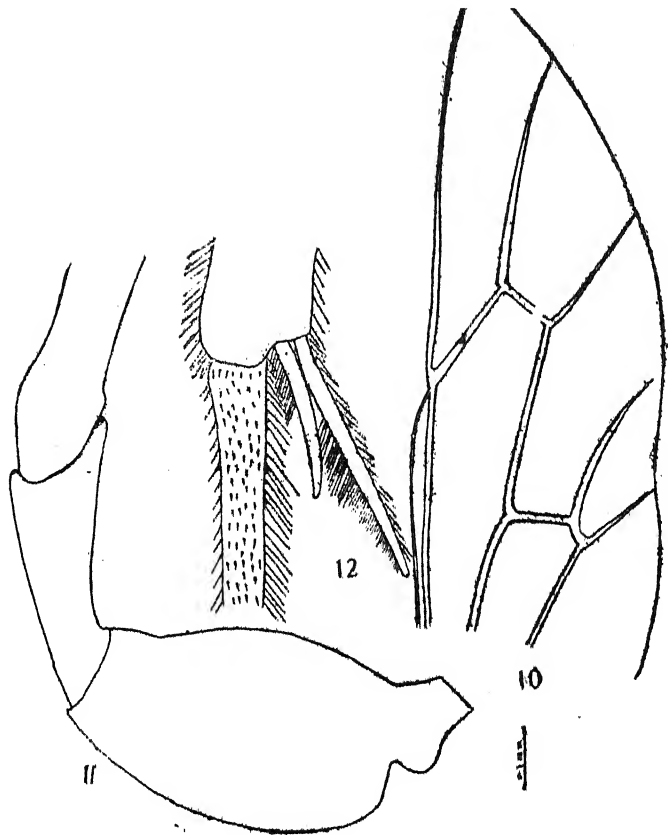
This species has very close resemblance to *silvaraji* R & K (5) in having three corneous marks but is easily distinguished by the differences in the wing venation, in the number of antennal segments and in the structure of the clypeus.

Tribe Ophionini

Stauropodoctonus indica, sp. nov.

♀ Body yellowish-brown, with black markings on thorax and abdomen. Head viewed from above transverse, slightly broader than thorax, twice as broad as long, ocelli prominent, in a triangle, interocellar space twice the ocellular and two-fifths the diameter of ocellus, posterior ocelli nearly confluent with the eye, interorbital space only half the width of head, vertex convex, smooth, pubescent, occiput excavate sparsely pubescent. Head viewed from front (fig. 21) broadly oval, frons reddish-yellow, flat with a median depression, face rugosely punctate, sparsely pubescent, reddish-brown, moderately convex, eyes naked and margined, clypeus nearly flat, distinctly marked out, pale yellow, rest as in face, mandibles dark reddish brown, apices black, antenna (figs. 22, 23 and 24) inserted above face, filiform, reddish-brown, finely pubescent, incomplete (43 segments only present), flagellate antennal segments cylindrical, scape (fig. 23) reddish-brown, nearly oval, length one and one-fifth the maximum width, sparsely hairy, pedicel (fig. 23) concolorous, broader than long (16:12). Head viewed from side oval, one and one-third times as long as thick, temples one-third the width of eye, gene one-seventh the length of eye.

Thorax reddish-brown, mesonotum closely finely pubescent, shallowly rugosely punctate with two median parapsidal furrows, scutellum convex, glabrous triangular,



Enicospilus indica, sp. nov. 10. Hind wing, 11. Hind coxa and trochanter, 12. Hind tibials pils, 13. Scape, pedicel and first flagellar segment, 14. Sixth to eighth antennal segments, 15. Forty to forty-two antennal segments and 16. Hind claw.

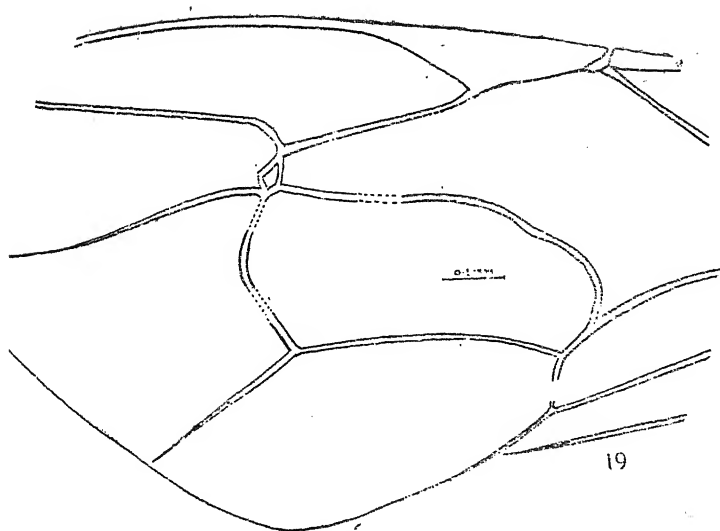
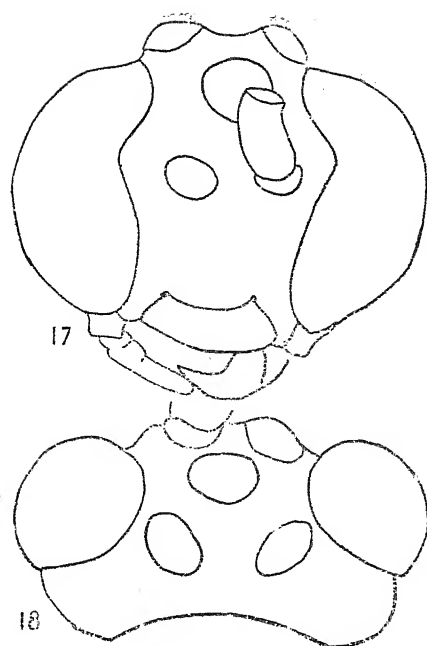


Fig. 17. Head front view and 18 . Head viewed from above of *Enicospilus indica* and
19. Fore wing of *Stauropodactonus ind ca*, sp. nov.

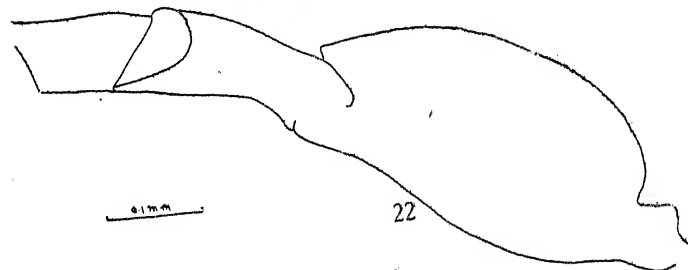
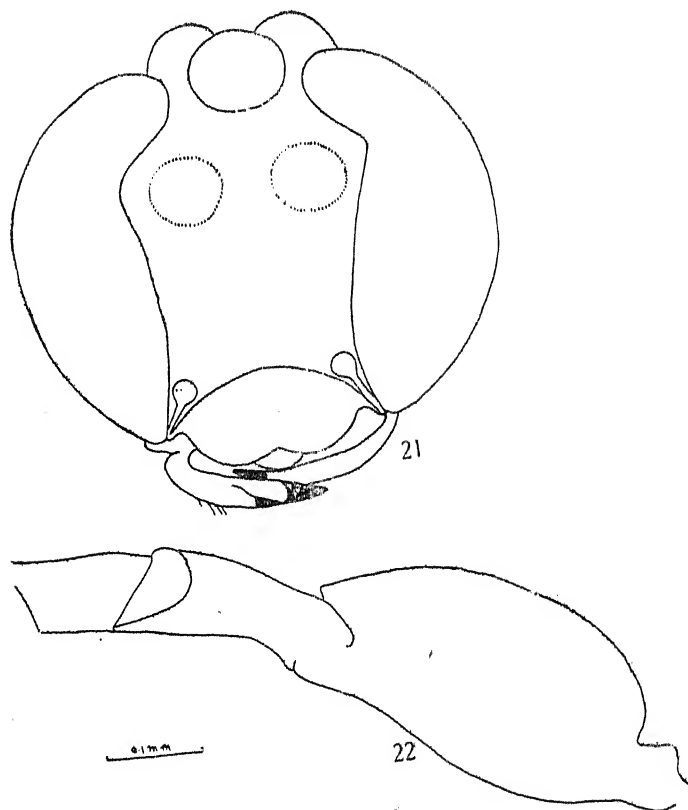
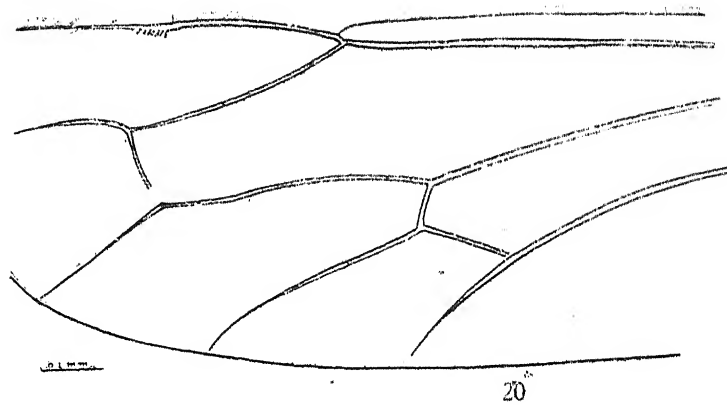


Fig. 20. Hind wing, 21. Head seen from front and 22. Hind coxa and trochanters of *S. indica*.

moderately convex, closely finely and shallowly punctate, finely pubescent, laterally margined, mesopleurae finely closely pubescent, rugosely punctate, tegulae blackish-brown; propodeum without an areola, finely pubescent, rugoso-punctate, with a distinct basal transverse carina, petiolar area of propodeum not distinctly marked off from the basal region, spiracle large, linear. Legs long, slender, reddish brown, covered with fine pubescence, hind leg largest and stoutest of all, coxa (fig. 22) smooth, pubescent, slightly longer than both trochanters combined, trochanter twice the trochanterellus; femur pubescent, longer than coxa and trochanters combined, tibia slender, length one and one-fourth the length of femur, covered with short pubescence, and with irregularly distributed small spines, tibial spurs (fig. 25) two, brown, unequal, longer spur about half the length of metatarsus, finely pubescent, very slightly and evenly curved, shorter spur only two-thirds the length of longer spur, finely pubescent, tarsal segments covered with fine pubescence, stiff setae and also spines, metatarsus a little over half the length of tibia, shorter than the rest of tarsal segments combined, with long stiff spines on the inner side, in addition to pubescence, claw pectinate (fig. 26), with a large number of thick teeth. Wings (figs. 19 and 20) large, membranous,

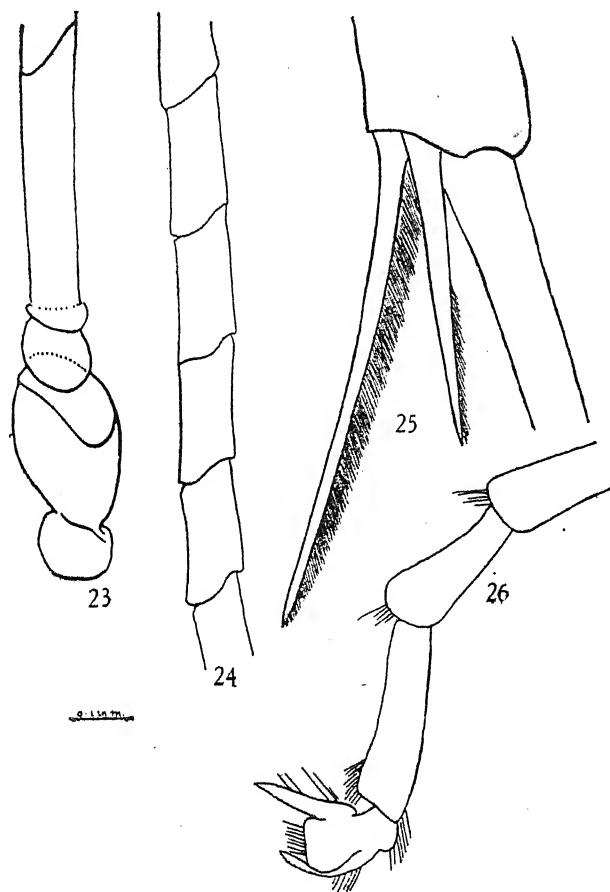


Fig. 23. Scape, pedicel and first flagellar segments, 24. Some middle antennal segments, 25. Hind tibial spurs and 26. Hind claw of *S. indica*.

two and three-fifths as long as broad, veins dark brown, except for the yellowish-brown unpigmented parts of M_{3+4} and M_2 , pterostigma yellowish brown, narrow, long, $SR + R_3$ distinctly curved, cell R_2 long, narrow, cell R_3 incomplete, with a short petiole, quadrate, issuing vein M_3 from middle, the latter curved, unpigmented basally as well as a little below its middle, M_{3+4} curved conspicuously and unpigmented at its apical third, cubital cell without corneous marks, rest of details and hind wing as in figures 19 and 20.

Abdomen longer than head and thorax combined, dark reddish-brown, finely and moderately covered with scanty and short pubescence, petiolate, strongly laterally compressed, terminal few tergites with dull black markings, first tergite long narrow, with keel-like lateral expansions at base, narrow basally, gradually becoming broader towards the apex, with a kind of a ridge at the basal one-third; second tergite nearly half of the length of first tergite, broad also compressed, covered with scanty fine pubescence, broader at apex than at base, with a lateral small circular spiracle in the basal half, terminal segments very much compressed, ovipositor and valvulae exerted.

Length ♀ 19 mm.

Holotype one ♀ on pin labelled "At light, Dining Table, S. N. Rao Coll., Ramdas-peth, Nagpur, 1956". Wings, legs and antenna mounted on slides.

This species has superficial resemblance to *orientalis* Morley (1) but is easily distinguished from it on account of the differences in the structure of the wing and thorax. The differences in the wing which are more striking are the absence of infumate area in the Radial cell and subapical part of first cubital cell.

Tribe *Cremastini*

Pristomerus (Pristomeridia) secunda (Morley)

1913. *Pristomeridia secunda*, Morley, *Faun. Brit. India, Hymen.*, 3:510.

1951. *Pristomeridia secunda*, Rao and Kurian, *Indian J. Ent.*, 13(1):26.

1953. *Pristomeridia secunda*, Rao, *Indian For. Rec.*, 8(8):192.

We have before us one ♀ labelled "On *Dia iscus*, Maharaj Bagh, S. N. Rao Coll., 14-2-1957, on wing, Nagpur" which is previously reported from Agra in Uttar Pradesh, Saugor in Madhya Pradesh and Wynaad from South India. This is the first record of this species from Nagpur (Vidarbha) Bombay State.

Tribe *Porizonini* (= *Campoplegini*)

Angetia fenestralis (Holmgren)

1829. *Campoplex majalis*, var. 4., Gravenhorst, *Ichn. Europe*, 3:464 ♀.

1858. *Limneria fenestralis*, Holmgren, *Svenska Vetensk. Akad. Handl.*, 2(2):59 ♂♀.

1878. *Limneria fenestralis*, Brischke, *Schr. Nat. Ges. Danzig.*, 6:150.

1885. *Limneria fenestralis*, Bridgman & Fitch, *Entom.*, p. 108 ♂♀.

1887. *Angetia fenestralis*, Thomson, *Opuscula Entomologica*, 11:156 ♂♀.

1913. *Angetia fenestralis*, Morley, *Faun. Brit. India, Hymen.*, 3:497 ♂♀.

1951. *Horogaster fenestralis*, Muesbeck, et al., U. S. Dept. Agric. Mongr. 2, p. 378.

1951. *Angetia fenestralis*, Rao and Kurian, *Indian J. Ent.*, 13(1):25 ♂.

We refer to this species one ♂ labelled "S. N. Rao Coll., Nagpur, On wing, 18-2-1957, Kitchan garden".

This species was previously recorded from Lyallpur (Pakistan), Mt. Abu (Rajasthan) and Agra (U. P.). This is the first record of this species from Nagpur (Vidarbha), Bombay State.

Subfamily *Mesochorinae*

Tribe *Mesochorini*

Pseudochorus kuriani Rao

1952. *Pseudochorus kuriani*, Rao, *Indian For. Rec.*, 8(8):195-198 ♂♀.

From among the collections under study is found one ♀ labelled "Maharaj Bagh, Nagpur, On mustard plant, S. N. Rao Coll., 14-2-1957" and one ♂ labelled "Kitchen Garden, S. N. Rao Coll., Nagpur, 18-2-1957" that are referred to this species.

This species was reported previously from Agra and is for the first time recorded here from Nagpur (Vidarbha) Bombay State.

Subfamily *Ichneumoninae*

Tribe *Ichneumonini*

Melcha nursei Cameron

1907. *Melcha nursei*, Cameron, *J. Bombay Nat. Hist. Soc.*, 17(3):593.

1951. *Melcha nursei*, Rao and Kurian, *Indian J. Ent.*, 13(1):33-36.

We refer to this species one ♂ labelled "S. N. Rao Coll., at Light, Aug. 1955, Ramdaspeth, Nagpur".

This is the first record of this species from Nagpur (Vidarbha) Bombay State.

Melcha ornatipennis Cameron

1907. *Melcha ornatipennis*, Cameron, *Ann. Mag. Nat. Hist. London*, 7:179.

1945. *Melcha ornatipennis*, Ahmed and Mathur, *Indian J. Ent.*, 7 (1 & 2):21-35.

1951. *Melcha ornatipennis*, Rao and Kurian, *Indian J. Ent.*, 13(1):36-38.

1953. *Melcha ornatipennis*, Rao, *Indian For. Rec.*, 8(8):213.

Labelled "On plant, S. G. Hardas, Aug. 1956, Nagpur" and "On wing, Dharmapeth, S. G. Hardas, Sept. 1956, Nagpur" are found two ♀♀ of this species which could be identified at sight by the characteristic white bands on the antenna and abdomen.

This is the first record of this species from Nagpur (Vidarbha), Bombay State.

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THE BOTANY OF COORG FORESTS

1. GENERAL

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Contributions to our knowledge on the forests of Coorg district are very scanty. The available references indicate that most of the work in this aspect has been carried on by the forest officers (Nanaya, 1949; Trimurti, 1955; Sumiah, 1957) though plants from this area have earlier been reported by workers like Somerson (1879), and Cameron (1894). Keeping in mind, the scope of work, studies on the floristics of this district were undertaken in hand in 1957. While the successional details and the composition of the communities under different vegetation types will be described later on, an attempt has been made in the present paper to lay out the broad features of vegetation. The paper thus presents in particular, the floristic composition of the different types of forests met with in Coorg.

The district of Coorg occupies a very prominent position along the humid tropic belt of the western ghats and is well known for its luxurient vegetation. With its high mountain ridges, narrow valleys and thickly wooded hills, this part of the country is situated between Lat. $11^{\circ} 55' N$ & $12^{\circ} 50' N$ and Long. $75^{\circ} 25' E$ & $76^{\circ} 14' E$ and has a total area of approx. 1600 Sq. miles. The district stretches to the maximum of 60 miles from Hemavati in the north to Devasibetta in the south, while its greatest breadth from Sampajee in the west to Fraserpeth in the east is about 40 miles. Towards north it forms a narrow area of about 12 by 6 miles which projects into the tableland of Mysore (Fig 1).

The southern boundry of the district is formed by the thickly wooded tracts of Wyanad and north Malabar (Kerala state), while on its north, northeast it joins the Tulu country or the district of south Kanara. The east of the district merges into the tableland of Mysore where for the most part, scrubby jungles of hardy, thorny species of *Zizyphus*, *Flacourtia indica*, *Gymnosporia* and others are met with.

TOPOGRAPHY

Coorg is hilly country and the range of irregular hills divides it into two parts; the uplands and the lowlands. The lowlands chiefly lie in the eastern and north eastern side and usually bear moist deciduous type as the best form of vegetation. The uplands are generally densely wooded and the climax form of vegetation in these areas is a typical wet evergreen type of forest, which has a similar physiognomy over most of the area with a different floristic composition. Mixed associations of evergreen species (*Mesua-Calophyllum*/*Dipterocarpus* type and others) are met with here.

At higher elevations, the summit of the hills are generally covered with grasses (*Themeda*, *Heteropogon* and others), scattered trees and woods in the hollows, through which flow various streams and rivulets. Such areas near Bagamandala range, present the characteristic shola-type appearance with vegetation condensed in pouches. Tree species of *Glochidion*, *Olea dioica* and others are often observed with *Wendlandia notoniana* and *Vernonia* sp., as common shrubs. Some of the prominent peaks

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offering a temperate type of climate with vegetation of *Rosa*, *Rubus*, *Anemone*, *Hypericum*, *Rhododendron* and others are :

| | |
|-------------|-------|
| Tadiandamol | 5725' |
| Puspagiri | 5620' |
| Kotebetta | 5375' |
| Brharmagiri | 5277' |

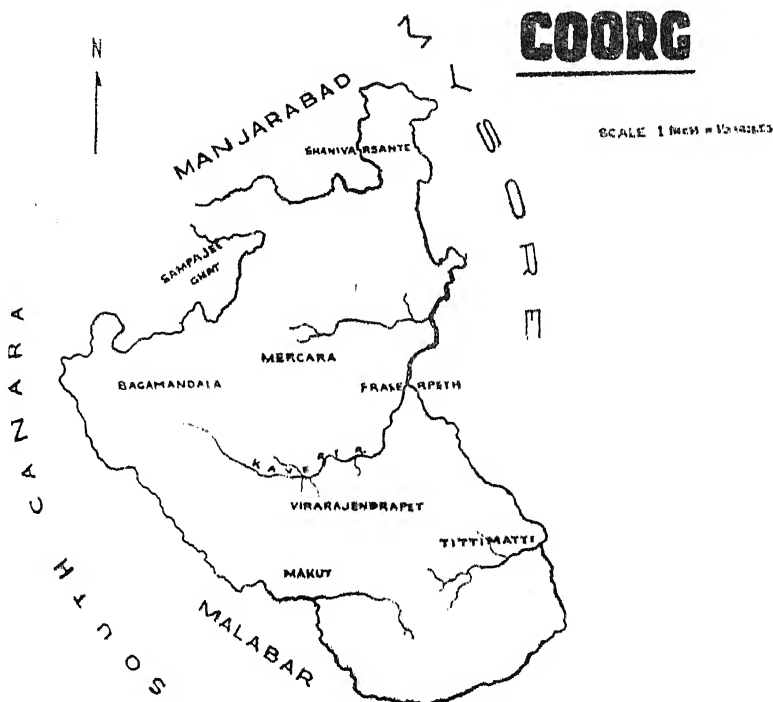


Fig. 1

DRAINAGE

The main drainage of the district is in easterly direction towards bay of Bengal, while the streams on the western slope follow a westerly course into the Indian ocean. The four main rivers of the district are : Kaveri, Barapole, Hemavati and Lakshmantirtha.

GEOLOGY

Geologically Coorg is formed of unclassified rocks *i.e.*, granites, gneiss & laterites. In the lowlands mainly claystone, limestone, micascists are met with. Near Mercara claystone or argillaceous schist of coarser variety, and at Bellur limestone have been reported to occur.

In addition to the old rock formation which provides the soil *in situ* by decomposition, alluvial deposits also occur along various streams and river beds. Such deposits usually support a good growth of Teak-Bamboo types.

The red soils derived from granite, which are also mixed with ferruginous deposits bear often the best developed evergreen forests in the district.

The rock usually is at considerable depth below the surface in the lowlands of the district which bear Teak & or Bamboo types. The soil here is fairly deep and black in colour. In the uplands, the soil is red, derived from granite, gneiss or laterite. The rock outcrops on surface and when the underlying substratum is a laterite, the hard honey-combed brownish red feature of this, can well be observed. Such a condition is chiefly seen along hillocks at higher elevations.

CLIMATE

The district offers a monsoonic-temperate type of climate. The distribution of rainfall per annum varies considerably from eastern to the southern and northern parts. The areas merging with the table-land of Mysore usually get a low rainfall while other towns in the eastern Coorg i. e., Kalhalla, Tittimati, Dubare, Nagarhole, Murkal, get a moderate amount of rainfall (upto 175 cms.) per annum. The differences in the amount of rainfall in the different areas of the district are given in Table I. Soil

TABLE No. 1

| Place | Rainfall in cm. | Vegetation type | Soil |
|-------------|-----------------|---|---|
| Hansur | 68 | Scrub forest | Hard, shallow, greyish |
| Fraserpeth | 93 | Scrub and dry Teak types dominate | Black soil |
| Murkal | 125 | Deciduous..... moist deciduous forests of Teak/Teak-Bamboo types. | Black soil, mostly alluvial |
| Tittimatti | 132 | | |
| Nagarhole | 163 | | |
| Kalhalla | 168 | | |
| Virajpeth | 263 | Disturbed vegetation near habitations: deciduous forests... semi-evergreen types in near about forests. | Black-Red soils |
| Mercara | 268 | | |
| Srimangala | 268 | | |
| Sampajee | 420 | Moist-deciduous, semi-evergreen, low evergreen types. | do |
| Kerike | 486 | Typical wet-evergreen type of forests dominate | Red soils, chiefly derived from granite, gneiss or laterite |
| Makut | 508 | | |
| Bagamandala | 550 | | |
| Watekolly | 551 | | |
| Pulingoth | 593 | | |

features are also indicated and apart, correlation of rainfall with the vegetation types is shown. Thus it is obvious that :

- a. the low rainfall areas of the district support scrubby type of forests (Hunsur) or at places dry-teak types with more of *Pterocarpus marsupium*.
- b. the moderate rainfall areas bear good Teak/Teak-Bamboo type forests (Kalhalla, Nagarhole, Tittimatti).
- & c. the high rainfall areas support the best form of vegetation i.e., Tropical wetevergreen type (Bagamandala, Kerike, Makut).

Now considering the soil features, in relation to climate and aspect, it is observed that :

- (i) the black soils usually support scrub-deciduous forests
-----Rainfall low
- (ii) the red soils generally bear evergreen types
-----Rainfall high

The rainfall patterns of the main towns are given in fig. 2.

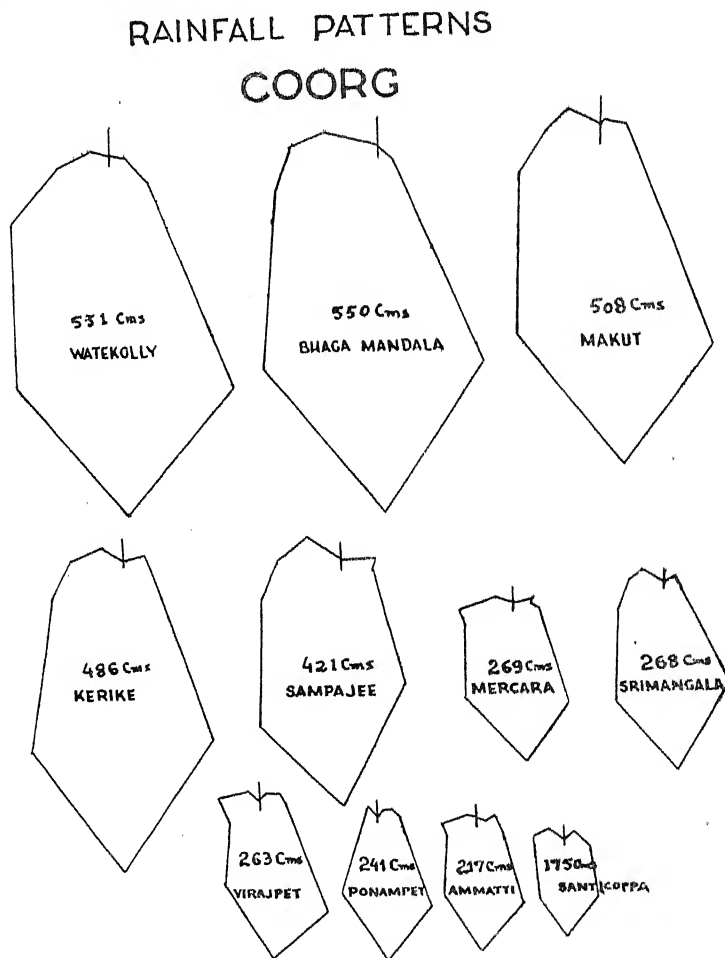


Fig. 2

Temperature shows more extremes (Max. 100°F; Min. 50°F) in the eastern Coorg and adjoining areas, while in the uplands it is very moderate (Max. 85°F; Min. 55°F). The maximum-minimum of temperature in relation to rainfall is indicated in Fig. 3., for Mercara.

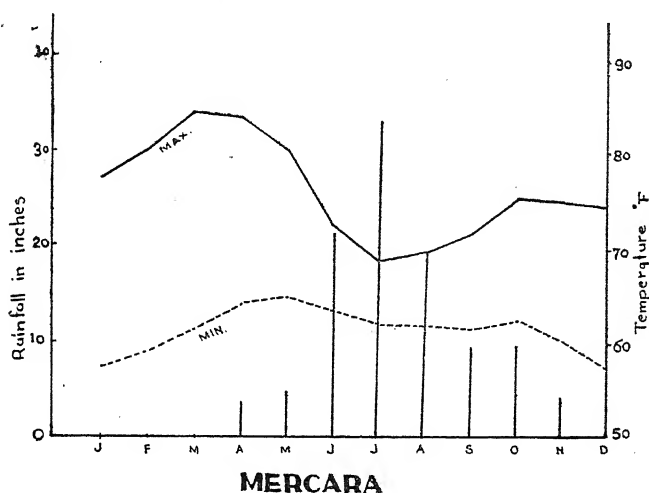


Fig. 3

FOREST TYPES

Based on the factors of the locality *i.e.*, rainfall, temperature and soil, three vegetation types are met with in Coorg, viz., evergreen, deciduous and scrub forests. There is however not very definite and clear demarcation in these types and so intermediate forms of vegetation are often to be seen in adjoining areas. Thus, between an evergreen forest there may be an interruption of a deciduous patch or in a deciduous forest a patch may contain a semi-evergreen or evergreen type of vegetation. However broadly, the evergreen type is more restricted to the high rainfall areas while low to moderate rainfall areas contain scrub or deciduous and moist-deciduous type of vegetation.

Evergreen type:

The wet-evergreen forests of the district are distributed over the long belt of Subramanya range which lies on the western ghats. These have been studied in the areas of Bagamandla, Kerike, Makut and Sampaji, which get a high rainfall of over 500 cms. per annum, a moderate temperature between 55°F to 85°F and are roughly situated 3000' above sea level. The density and the composition of the vegetation varies to some extent due to multieffect of the various factors like soil, drainage conditions, steepness and wind exposure etc., though the general evergreen form is retained over all these areas.

In these forests, the vegetation occurs in stratification and about 3-4 layers can be demarcated on height basis; usually the top canopy or the 1st storey attains a height of 100'-125' though sometimes it may exceed 150'. It is composed of straight tall trees which have a clean bole upto 8-10 metres. Most of the trees (*Tetrameles*

nudiflora, *Elaeocarpus tuberculatus*, *Eipterocarpus turbinatus*, *Dysoxylum malabaricum*, *Diospyros microphylla* and others) develop huge buttresses giving a magnificent appearance. This story is followed by the second layer (50'-75') which has a mixed composition with a good number of trees tending to reach the top storey level (*Myristica attenuata*, *Cinnamomum zeylanicum*, *Acrocarpus fraxinifolius*, *Elaeocarpus tuberculatus*, *Diospyros* sp. and others).

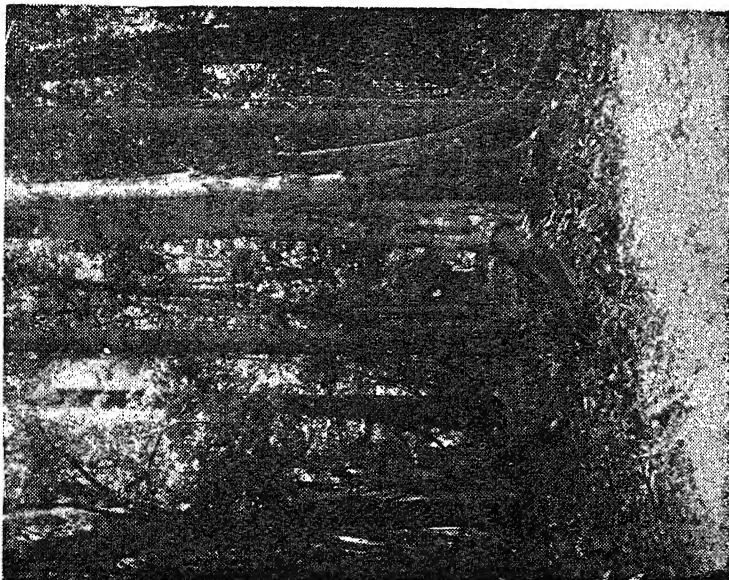
The third storey is generally of low trees, shrubs and climbers, but seldom in forests where the top storey exceeds 150', the low trees retain a characteristic stratification and shrubs and climbers form a distinct layer underneath these. The growth of the lianes like *Gnetum ula* and *Anistrocladus heyneanus* is dense and often forms impenetrable thickets. *Calycopteris floribunda* usually occupies forest fringes. This is followed by the ground flora which constitutes the lowest height layer.

The forests usually are composed of mixed communities with three or more codominants. The following species are met with :—

- (a) Top layer or first storey species : variable in height, usual 100'-125'. Occasionally exceeding 150'.

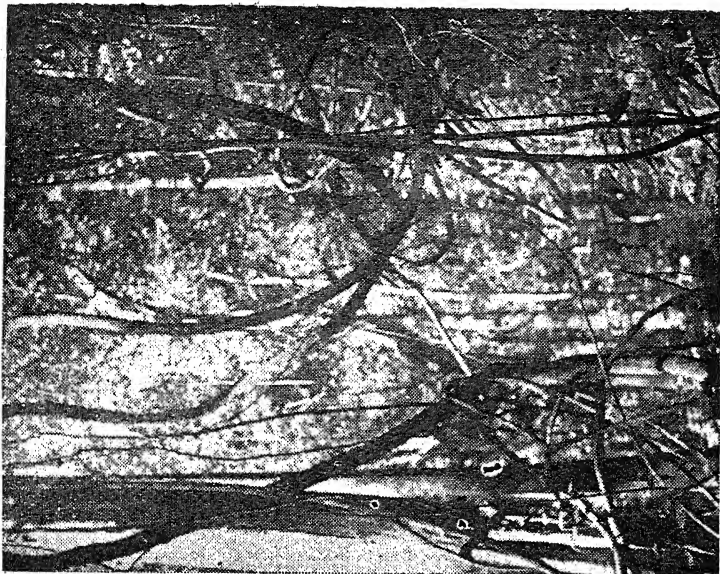
| | |
|--|--------------------------------|
| <i>Acrocarpus fraxinifolius</i> | <i>Albizzia stipulata</i> |
| <i>Antiaris toxicaria</i> | <i>Artocarpus hirsuta</i> |
| <i>Beilschmiedia fagifolia</i> | <i>Bischofia javanica</i> |
| <i>Calophyllum</i> sp. | <i>Canarium strictum</i> |
| <i>Caryota urens</i> | <i>Cinnamomum zeylanicum</i> |
| <i>Dipterocarpus turbinatus</i> | <i>Diospyros</i> sp. |
| <i>Donella roxburghii</i> | <i>Dysoxylum malabaricum</i> |
| <i>Elaeocarpus tuberculatus</i> | <i>Euphoria longana</i> |
| <i>Evodia roxburghiana</i> | <i>Ficus</i> sp. |
| <i>Garcinia</i> sp. | <i>Holigarna grahamii</i> |
| <i>Hopea parviflora</i> | <i>Hopea wightiana</i> |
| <i>Knema attenuata</i> | <i>Lepisanthes tetraphylla</i> |
| <i>Litsea</i> sp. | <i>Lophopetalum wightianum</i> |
| <i>Macaranga peltata</i> | <i>Mesua ferrea</i> |
| <i>Myristica</i> sp. | <i>Palaquium ellipticum</i> |
| <i>Pithecolobium bigeminum</i> | <i>Polyalthea fragrans</i> |
| <i>Strychnos nux-vomica</i> | <i>Syzygium</i> sp. |
| <i>Tetrameles nudiflora</i> | <i>Trewia nudiflora</i> |
| <i>Vateria indica</i> , with less prominent species like, <i>Vitex</i> sp. | |
| <i>Lagerstroemia speciosa</i> | <i>Michelia champaca</i> |

PHOTO 1



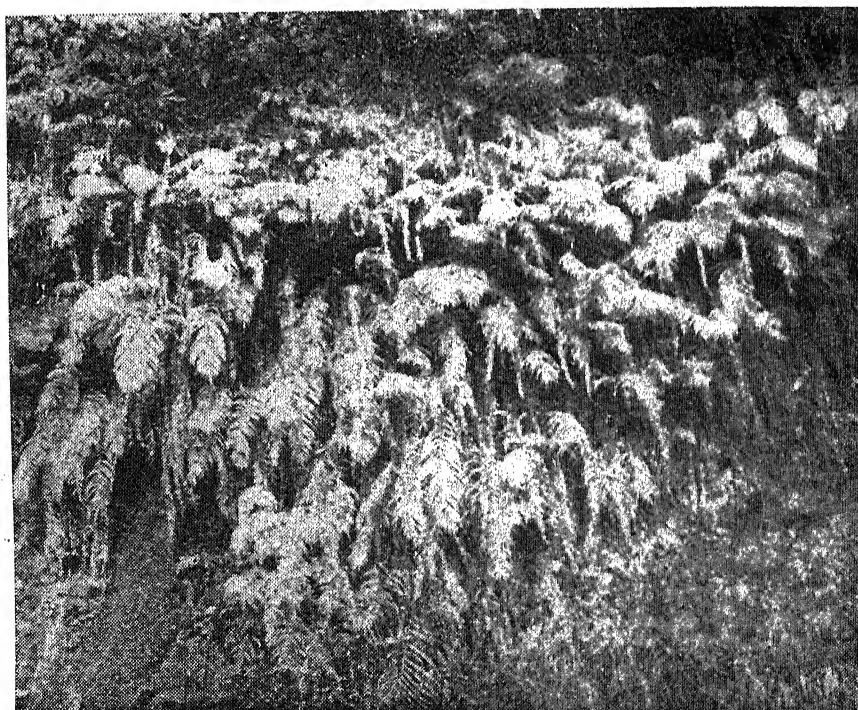
View of an evergreen forest with buttressed trees
of *Tetramelis macrostoma* and others.

PHOTO 2



A tangle of lianes (*Gnetum ula* and others) in
evergreen forests.

PHOTO 3



Dense growth of *Angiopteris evecta*, *Blechnum* and others along edges of evergreen forests.

- (b) Second storey : is usually less prominent with a mixture of top storey species :

| | |
|---------------------------------|-----------------------------|
| <i>Acrocarpus fraxinifolius</i> | <i>Actinodaphne</i> spp. |
| <i>Agalia roxburghiana</i> | <i>Alstonia scholaris</i> |
| <i>Aporosa lindleyana</i> | <i>Artocarpus lakoocha</i> |
| <i>Bischofia javanica</i> | <i>Carallia</i> sp. |
| <i>Cinnamomum zeylanicum</i> | <i>Diospyros</i> sp. |
| <i>Elaeocarpus tuberculatus</i> | <i>Euphoria longana</i> |
| <i>Flacourtia montana</i> | <i>Hardwickia pinnata</i> |
| <i>Hemicyclia alata</i> | <i>Holigarna arnotiana</i> |
| <i>Hydnocarpus wightiana</i> | <i>Knema attenuata</i> |
| <i>Litsea</i> sp. | <i>Machilus macrantha</i> |
| <i>Olea dioica</i> | <i>Strombosia ceylanica</i> |
| <i>Strychnos nux-vomica</i> | <i>Terminalia</i> sp. |
| <i>Tetrameles nudiflora</i> | <i>Vitex altissima</i> |
| <i>Xanthophyllum flavescens</i> | <i>Xylin xylocarpa</i> |

- (c) Third storey is composed of small trees exceeding 20' in height.

| | |
|-----------------------------|---------------------------------|
| <i>Aporosa lindleyana</i> | <i>Callicarpa tomentosa</i> |
| <i>Canthium dicoccum</i> | <i>Flacourtia montana</i> |
| <i>Ixora brachiata</i> | <i>Ixora nigricans</i> |
| <i>Lancium anamalayanum</i> | <i>Leea indica</i> |
| <i>Memecylon</i> sp. | <i>Psychotria</i> sp. |
| <i>Schefflera</i> sp. | <i>Xanthophyllum flavescens</i> |

- (d) Fourth storey is composed of shrubs and climbers which grow in abundance in these forests often forming impenetrable thickets.

| | |
|------------------------------|--------------------------------|
| <i>Allophylus serratus</i> | <i>Anastrocladus heyneanus</i> |
| <i>Arenga wightii</i> | <i>Asparagus racemosus</i> |
| <i>Calamus</i> sp. | <i>Calycoternis floribunda</i> |
| <i>Chasalia curviflora</i> | <i>Cissus</i> sp. |
| <i>Clerodendron</i> sp. | <i>Cyclea</i> sp. |
| <i>Entada phaseoloides</i> | <i>Gnetum ula</i> |
| <i>Hemidesmus indicus</i> | <i>Ixora nigricans</i> |
| <i>Jasminum rottlerianum</i> | <i>Melanthesea turbinata</i> |
| <i>Murraya koenigii</i> | <i>Naravelia zeylanica</i> |

Piper nigrum
Smilax zeylanica
Vepris bilocularis

Pothos scandens
Webera corymbosa
Zizyphus sp.

The lowest layer is of ground flora species which are chiefly composed of seedlings of trees, shrubs etc. Amongst other plants *Curcuma*, *Costus* and other Zingiberaceae with a good growth of ferns like *Pteris* sp., *Gymnopteris* sp., *Stenoloma* sp. and others are commonly noted.

The fringes of these forests generally support species like *Lantana*, *Jasminum*, *Calycopteris*, *Streblus asper* and *Carvia callosa*.

Cryptogamic flora is rich and many species of *Agarics* and *Polypores* are often observed on dead decaying woods. Musci like *Micrometrium* and others are also common. The epiphytic flora is well represented by orchids and ferns. Of the common orchids *Aerides*, *Dendrobium* sp., *Pholidota imbricata*, *Acampe wightiana*, *Oberonia* sp., are met with. Of the epiphytic ferns *Pleopeltis* spp., are generally observed, the most common being *Pleopeltis membranacea*. *Drynaria quercifolia* is usually found on the outskirts of these forests often on *Ficus* sp.

Deciduous forests :—

The deciduous forests of the district are mainly distributed along the eastern belt, extending over the Fraserpeth range south of Mercara-Fraserpeth and whole of Tittimatti, Kalhalla and Nagarhole ranges east of Mercara-Manantody. All these areas roughly lie between an elevation of 2000' to 3000' rarely above this when they tend to progress into semi-evergreen type of vegetation.

The moist deciduous forests are best developed in the eastern and central regions which get comparatively a moderate rainfall of 150-200 cms. per annum. In more moist localities this type shows a mixed composition with semi-evergreen species like *Tabernaemontana heynana*, *Xyia xylocarpa*, *Callicarpa tomentosa* and others. The vegetation along the eastern belt of the district adjoining Mysore is of drier type of deciduous forests. This has been studied in the areas of Hunsur and Fraserpeth ranges which get a low rainfall of about 100 cms. The chief associates of these forests are *Anogeissus latifolia*, *Terminalia tomentosa*, *Madhuca indica*, *Diospyros melanoxylon*, and others, while the moist deciduous type is recognised by the presence of bamboos i.e., *Bambusa bambos*, *Dendrocalamus strictus*, with other species like *Dillenia pentagyna*, *Azina cordifolia*, *Emblia officinalis* and *Grewia tiliifolia*. *Tectona grandis* is present in both the types though in the former it has a poor stunted scrubby growth.

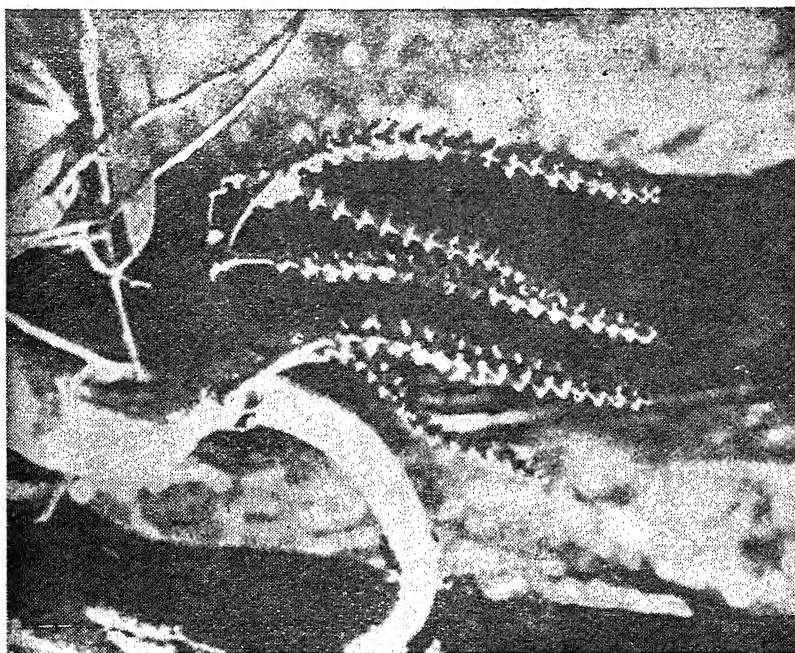
These forests do not show any characteristic stratification. The trees attain a height of 50'-75' and the canopy is never too thick. Buttressed nature of trees which is characteristic of the wet-evergreen type is usually uncommon, mostly missing here. Thick growth of climbing species met within these forests exceeds those of evergreen type.

The chief tree species from these forests are :

Adina cordifolia
Bauhinia sp.
Butea monosperma

Albizzia sp.
Bridelia squamosa
Dalbergia latifolia

PHOTO 5



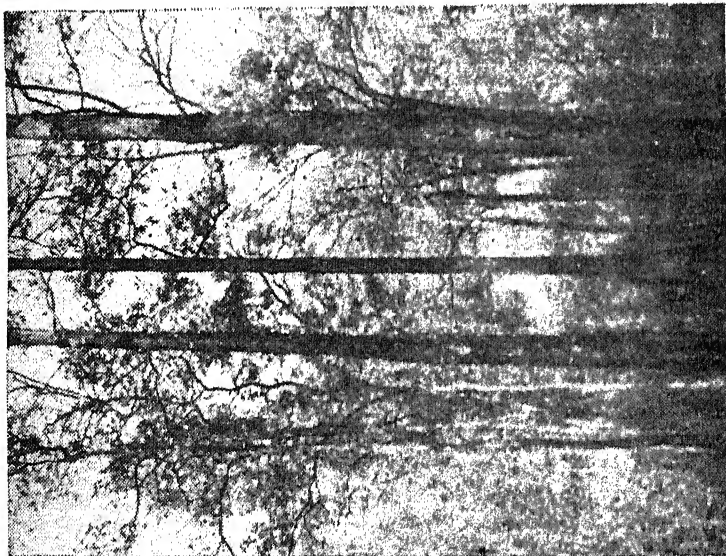
Gnidium ulu, the common climber in evergreen forests.

PHOTO 4



Costus speciosus, common undergrowth in evergreen forests.

PHOTO 6



Mixed deciduous forest with Teak, *Actia coriifolia*, *Lagerströmia lanceolata*, *Dillenia pentagyna* and others with an undergrowth of bamboos.

PHOTO 7



Close view of Bamboo clumps (*Dendrocalamus strictus*).

Dillenia pentagyna

Ehretia laevis

Ficus sp.

Gmelina arborea

Kydia calycina

Lannea grandis

Pterocarpus marsupium

Saccopetalum tomentosum

Sterculia sp.

Terminalia tomentosa

Terminalia chebula

Zanthoxylum rhetsa

Diospyros montana

Emblica officinalis

Garuga pinnata

Grewia tiliaefolia

Lagerstroemia lanceolata

Mallotus philippensis

Schleichera oleosa

Shorea tolura

Tectona grandis

Terminalia paniculata

Xylia xylocarpa

Amongst small trees and shrubs following species are more common

Carissa congesta

Colebrookia oppositifolia

Meyna laxiflora

Lantana camara

Randia brandisii

Zizyphus sp.

Callicarpa tomentosa

Holarrhena antidysenterica

Murraya koenigii

Leea indica

Solanum sp.

The common climbers are :

Asparagus racemosus

Cissus sp.

Diploclisia glaucescens

Calycopteris floribunda

Cryptolepis buehanani

Entada phaseoloides

The undergrowth consists of :

Carvia callosa

Desmodium sp.

Moghania strobilifera

Urena lobata

Crotalaria sp.

Mimosa pudica

Sida rhombifolia

With grasses like *Themeda*, *Apluda*, *Eragrostis* and *Oplismenus* sp. *Bambusa bambos* and *Dendrocalamus strictus* are the common bamboos met within these forests.

Scrub forests :—

The scrub type too is distributed along the eastern belt of Coorg where a vegetation of dry deciduous species is met with.

Rainfall in the areas which support scrub forests is very low (Hunsur, 68 cms. ; Fraserpeth, 93 cms.). The vegetation chiefly consists of thorny species with a few stunted, crooked and malformed trees.

The chief components of the scrub forests are :—

| | |
|-----------------------------------|---------------------------|
| <i>Acacia catechu</i> | <i>Argyrea cuneata</i> |
| <i>Balanites aegyptiaca</i> | <i>Capparis</i> sp. |
| <i>Carissa congesta</i> | <i>Cassia auriculata</i> |
| <i>Cipadessa bacifera</i> | <i>Dodonaea viscosa</i> |
| <i>Erythroxylon</i> sp. | <i>Euphorbia</i> sp. |
| <i>Flacourtia indica</i> | <i>Fluggea</i> sp. |
| <i>Gardenia</i> sp. | <i>Gymnosporia</i> sp. |
| <i>Ixora parviflora</i> | <i>Lantana camara</i> |
| <i>Pavetta indica</i> | <i>Randia brandisii</i> |
| <i>Rhus mysorensis</i> (uncommon) | <i>Soyimida febrifuga</i> |

The tree species commonly observed in these forests are :

| | |
|--------------------------------|--------------------------------|
| <i>Anogeissus latifolia</i> | <i>Bauhinia racemosa</i> |
| <i>Bridelia squamosa</i> | <i>Buchanania lanzan</i> |
| <i>Careya arborea</i> | <i>Cassia fistula</i> |
| <i>Chloroxylon swietenia</i> | <i>Cochlospermum gossypium</i> |
| <i>Diospyros melanoxylon</i> | <i>Lagerstroemia</i> sp. |
| <i>Pterocarpus marsupium</i> | <i>Seimecarpus anacardium</i> |
| <i>Sterospermum xylocarpum</i> | |

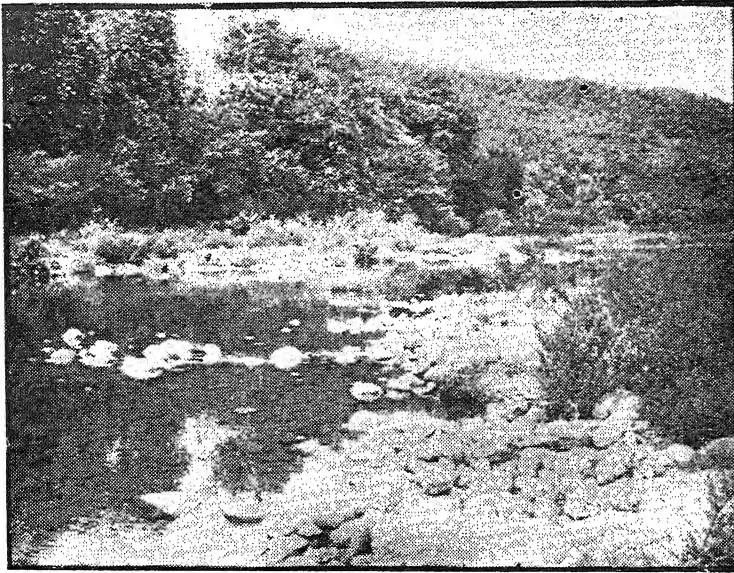
Boswellia serrata may also be observed at some places but is rather uncommon. Sandalwood (*Santalum album*) seems to prefer these open jungles with scrubby associates.

Amongst the undergrowth grasses like *Apluda varia*, *Aristida* sp., *Eragrostis uniloides*, *Heteropogon contortus*, *Oplismenus* sp., and *Setaria glauca*, with herbs like *Andrographis* sp., *Blepharis* spp., *Polycarpea argentea*, *Clerodendron serratum*, *Crotalaria* sp., *Alysicarpus*, *Heylandia*, *Indigofera* spp., with hardy undershrubs of *Barleria*, *buxifolia* are met with.

Features of the flora in areas surveyed :

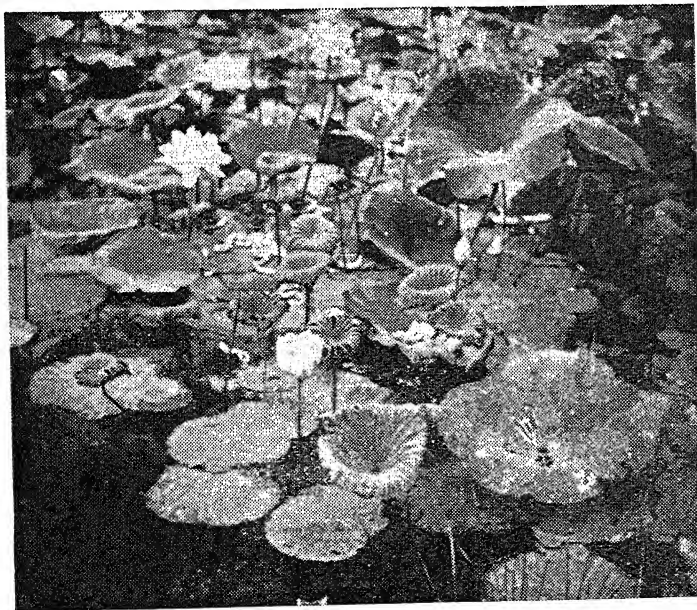
The areas of Hunsur, Kalhalla, Nagarholle. and Tittimatti have been explored in the low rainfall zone of the district and the deciduous forests which predominate in these areas, studied. In the high rainfall zone, the areas of Mercara, Sampajee, Makut, Bagamandala, and Keriki have been surveyed so far and the wet-evergreen type of vegetation studied to a considerable detail.

PHOTO 8



Riverian vegetation near Makut (*Phyllanthus lawii*-*Homonola* mixed type).

PHOTO 9



Close view of *Nelumbium* community.

In all 700 plant species have been collected of which over 400 are trees, while the rest have a good proportion of shrubs, climbers and herbs. The tree species belong to families like Annonaceae, Guttiferae, Meliaceae, Sapindaceae, Anacardiaceae, Leguminosae, Combretaceae, Rubiaceae, Myrtaceae, Ebenaceae, Myristicaceae, Lauraceae, Euphorbiaceae, Urticaceae and others. The climbers are well represented by families like Ranunculaceae, Menispermaceae, Olacaceae, Vitaceae, Leguminosae, Araliaceae, Oleaceae, Myrsinaceae, Apocynaceae, Asclepiadaceae, Loganiaceae, Convolvulaceae, Piperaceae, Dioscoreaceae and others. *Gnetum ula*, *Anastrodadus heyneanus*, *Calycopterus floribunda*, *Artabotrys zeylanicus* are the common climbers in evergreen types.

The herbacious plants belong to Geraniaceae, Malvaceae, Tiliaceae, Leguminosae, Rubiaceae, Labiatae, Amarantaceae, Cyperaceae, Gramineae, and others *i.e.*, Begoniaceae, Ficoideae, Umbelliferae, Gentianaceae, Scrophulariaceae, Acanthaceae, Polygonaceae, Liliaceae, Commellinaceae, Scitamineae, Eriocaulaceae. Epiphytic flora chiefly consists of ferns like *Drynaria quercifolia*, *Pleopeltis* spp. and a good number of orchids *i.e.*, *Pholidota imbricata*, *Bulbophyllum*, *Dendrobium* spp. *Aerides* spp. *Liparis* spp. *Oberonia* spp. and others. Amongst other plants, species of *Loranthus* are commonly observed.

The hydrophytic vegetation is composed of spp. like *Nymphaea*, *Nelumbium*, *Limnanthemum* and others. *Homonoia* spp. *Polygonum glabrum*, *Phyllanthus lawii*, *Ludwigia parviflora*, *Jussiaea suffruticosa* are often observed along stream beds.

SUMMARY

The paper presents in brief the observations of the author on the forest botany of Coorg district, studies on which are being carried on since 1957.

The distribution of the principal vegetation types is discussed and the floristic composition of the forests studied in different areas of the district described. The account of 700 species collected by the author, based on Benthum-Hooker system will be presented later on. Shola-forests will also be described separately.

ACKNOWLEDGEMENTS

The author expresses his grateful thanks to Dr. J. C. Sen Gupta, Dr. G. S. Puri, and to the Regional Botanist, Botanical Survey of India, Poona, for their needful guidance. He is equally grateful to C. S. I. R. authorities for the provision of a Junior Research Fellowship during the tenure of which, these studies have been made.

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DISTRIBUTION OF NITROGEN IN X-RAY PROGENIES OF *TRITICUM AESTIVUM* L.* DURING GROWTH

By

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INTRODUCTION

Sahasrabuddhe (1928) cited investigations carried out to correlate nitrogen distribution between the different parts of the plant at specified physiological stage and also the net nitrogen absorption to determine definitely the period at which nitrogenous additives could be applied for better responses. Singh (1939) in an account of a discussion on the absorption of salts by plants, recorded "definite and substantial losses of certain constituents notably potassium, nitrogen and calcium." He further stressed that "the loss is more or less concurrent with the migration of the same constituents into developing heads." The observance of the decrease of nitrogen and protein content in the vegetative parts, with the advance in age was also noted by Neidig and Snyder (1924) and Verma (1949). Woodford and McCalla (1936) showed that the composition of wheat plants (Reward and Red Bobs) grown on the black and gray soils of Alberta was influenced by the differences in soil and variety.

Neidig and Snyder (1922) also emphasized the importance of the influence of moisture on the protein content of wheat. These workers also showed that under field conditions, a high moisture content properly distributed during the growing season in an average soil produced a bumper wheat crop with a low protein content. Labov (1951) working on two varieties of oats observed a reduction of crude protein with increasing water applications.

There is some evidence on record to show the inhibition of enzymatic activity and denaturation of protein by the respective treatments with small and high doses of X-rays, but there is little data showing the behaviour of the effect of X-rays being transmitted through the progenies specially in relation to the protein fraction (Zirkle *et al.*, 1954).

The present work was undertaken in order to elucidate the fate of total nitrogen in the life-cycle of wheat plants NP₅₂ and its three new X-ray strains. The influence of field moisture on the crude protein content of these strains was also studied alongside.

EXPERIMENTAL

Wheat strains† NP₅₂, R₁, R₇ (Sarojini) and R₉ (Vijaya) were selected for the purpose of this study. Seeds were sown in randomized blocks. For observations plants

* Used originally for bearded spring wheat only, but now regarded as synonymous with and having priority over *T. vulgare*.

† NP₅₂ was obtained as a result of a cross between Pusa 6 and Punjab 9. R-strains (R=Ranjan) were developed by X-ray treatment of NP₅₂ seedlings for varying periods in the Department of Botany, University of Allahabad (Pugh, 1945).

were selected at random, cut just above the soil surface and brought to the laboratory in sampling bags. Nitrogen estimations of these plants were made at their physiological stages of growth, viz., tillering, pre-flowering, flowering, milk and dough. As the ears developed, the determinations were made for the foliage part and the ear-heads of the plants separately.

Gunning and Hibbard's method (Piper, 1947) was followed for the determinations of total nitrogen. Crude protein fraction of the plant material was obtained by multiplying the values for total nitrogen with the factor 6.25.

EXPERIMENTAL FINDINGS

Vegetative shoot. Total nitrogen percentage showed an increase from tillering to the preflowering stage but declined thereafter. Considering the variance in nitrogen values of the different strains for the physiological stages under observation, maximum percentage was recorded at tillering, milk and dough stages by R₁ plants; at preflowering by R₇ and at flowering by R₉. The minimum percentage of total nitrogen was noted in NP₅₂ plants at all the stages except at tillering and preflowering when R₇ and R₉ respectively were found to have lower values (Table I).

TABLE I. Total nitrogen and crude protein content of wheat at successive stages in the life-cycle
(Average percentage, oven dry basis)

| Physiological stages (days) | Strains | Vegetative shoot | | Inflorescence | | Tops | |
|--------------------------------|------------------|-------------------|------------------|-------------------|------------------|-------------------|------------------|
| | | Total Nitrogen | Crude Protein | Total Nitrogen | Crude Protein | Total Nitrogen | Crude Protein |
| Tillering (30) | NP ₅₂ | 0.374 | 2.1500 | ... | ... | 0.374 | 2.1500 |
| | R ₁ | 0.637 | 3.9812 | ... | ... | 0.637 | 3.9812 |
| | R ₇ | 0.259 | 1.6187 | ... | ... | 0.259 | 1.6187 |
| | R ₉ | 0.472 | 2.9531 | ... | ... | 0.472 | 2.9531 |
| Pretlowering (50) | NP ₅₂ | 1.470 | 9.1875 | 1.190 | 7.4375 | 2.660 | 16.6250 |
| | R ₁ | 1.540 | 9.6250 | 1.260 | 7.7750 | 2.800 | 17.4000 |
| | R ₇ | 1.680 | 10.5000 | 0.840 | 5.2500 | 2.520 | 15.7500 |
| | R ₉ | 1.400 | 8.7500 | 1.190 | 7.4375 | 2.590 | 16.1875 |
| Flowering (80) | NP ₅₂ | 0.970 | 6.0625 | 1.230 | 7.6875 | 2.200 | 13.7500 |
| | R ₁ | 1.230 | 7.6875 | 1.340 | 8.3750 | 2.570 | 16.0625 |
| | R ₇ | 1.130 | 7.0625 | 1.210 | 7.5625 | 2.340 | 14.6250 |
| | R ₉ | 1.280 | 8.0000 | 1.230 | 7.6875 | 2.510 | 15.6875 |
| Milk (90) | NP ₅₂ | 0.840 | 5.2500 | 1.680 | 10.5700 | 2.250 | 15.8200 |
| | R ₁ | 1.330 | 7.3125 | 1.890 | 11.8125 | 3.220 | 19.1250 |
| | R ₇ | 0.980 | 6.1250 | 1.610 | 10.0625 | 2.590 | 16.1875 |
| | R ₉ | 1.190 | 7.4375 | 1.540 | 9.6250 | 2.730 | 17.0625 |
| Dough (110) | NP ₅₂ | 0.810 | 5.0625 | 1.980 | 12.3750 | 2.790 | 17.4375 |
| | R ₁ | 0.988 | 6.1760 | 1.650 | 10.3125 | 2.638 | 16.4805 |
| | R ₇ | 0.860 | 5.3750 | 1.820 | 11.3750 | 2.680 | 16.7500 |
| | R ₉ | 0.970 | 6.0625 | 1.860 | 11.6250 | 2.830 | 17.6875 |

Inflorescence. The percentage of total nitrogen in ears was found to increase gradually after the preflowering stage irrespective of differences in the strains. Maximum nitrogen percentage was recorded by R_1 at all the stages except at dough when NP_{52} exceeded. The maximum total nitrogen percentage was, however, recorded by R_7 at preflowering and flowering, R_9 at milk and R_1 at dough (Table I).

It was found that as the total nitrogen percentage and crude protein in the vegetative tops decreased the percentage of total nitrogen in the ear-heads increased. Nitrogen was found to decrease at flowering after which the plants recorded a general rise except in the case of R_1 (Table I). The maximum percentage of crude protein was noted in R_9 at the dough stage and in R_1 at tillering, preflowering, flowering and milk. The minimum percentage of the same was, however, observed in R_7 at tillering and preflowering, in NP_{52} at flowering and milk and in R_1 at dough (Table I).

DISCUSSION

Crude protein percentage of the foliage of plants (tops less ear) was highest at the preflowering stage diminishing gradually to the lowest at dough while the reverse held true in the case of ear-heads. This opposite trend and behaviour exhibited by the ear-heads in contrast to the vegetative parts of the plant body was suggestive of the gradual transference of nitrogenous food material from the vegetative to the reproductive parts with the approach of the ripening stage. Translocation of this kind was also noted by Maskell and Mason (1930) in the cotton plant. Sircar and Sen (1941), in their studies on the physiology of rice, observed increase of total nitrogen as well as of protein nitrogen in the ear-heads. Sen (1946) also noted in the last two leaves of rice which emerged during the reproductive stage, a fall of protein content, and considered this to be due to the translocation of the nitrogenous products to the rapidly growing ear. This may be, as Sen (1946) and Petrie (1937) observed, due to the formation of young leaves and the emergence of inflorescences "which act like sink and increase the rate of translocation of nitrogen from the leaves below." This translocation was found to be highest in NP_{52} and lowest in R_1 .

The rate of increase of total nitrogen between the tillering and the preflowering stage was maximum in the tops (whole plant less root) while the minimum recorded was between the milk and dough stages. A sudden fall in the plant nitrogen at flowering was exhibited by all the plants; it is more in NP_{52} and less in the R-strains which is suggestive of some effect of X-rays on the protein dynamics at the time of seed formation. It seems that the yield of X-ray strains was adequately influenced by this fall of crude protein and that the time of X-ray exposure to seeds from which the plants grew also influenced the measure of this fall. It may be noted that among the R-strains this fall is negatively correlated with the yield of grains (Table V). The disturbance in the equilibrium of the crude protein status between the preflowering and the flowering stages of the X-ray progenies may, therefore, be held derogatory to the yield of their grains. According to Deleano (1936) a large proportion of the mineral elements and nitrogen in the foliage of trees is drained into the stems and roots before abscission of the leaves takes place. A perusal of tables III and IV depicts the sudden fall in the number of leaves and tillers from flowering till the end of the life-cycle. The fall immediately after the preflowering stage which recorded the maximum leaf number seems to have effected the immediate transference of the mineral elements and nitrogen to the stems and roots. In the subsequent stages, this fall did not affect the plant

TABLE II. Variations in moisture content of soil
supporting wheat strains

(Average percentage, over dry basis)

| Physiological stages (days) | Strains | Moisture Content |
|--------------------------------|------------------|------------------|
| Tillering (30) | NP ₅₂ | 10.808 |
| | R ₁ | 10.197 |
| | R ₇ | 9.713 |
| | R ₉ | 8.052 |
| Preflowering (50) | NP ₅₂ | 7.500 |
| | R ₁ | 8.800 |
| | R ₇ | 9.240 |
| | R ₉ | 9.300 |
| Flowering (80) | NP ₅₂ | 9.070 |
| | R ₁ | 8.570 |
| | R ₇ | 9.000 |
| | R ₉ | 9.200 |
| Milk (90) | NP ₅₂ | 5.660 |
| | R ₁ | 5.060 |
| | R ₇ | 4.960 |
| | R ₉ | 5.740 |
| Dough (110) | NP ₅₂ | 3.638 |
| | R ₁ | 2.530 |
| | R ₇ | 2.450 |
| | R ₉ | 2.791 |

nitrogen (entire tops) but instead due to the intensity of milk formation within the developing seeds was diverted to the ears (Fig. II). These results are in concurrence with that of Deleano (1936) for the flowering stage alone though not for the subsequent ones. This difference in behaviour might be due to the variance in the plant material put under test in the two investigations since the change in the nature of the plant has a remarkable effect on its metabolic processes (Singh, 1958).

Gradual depletion of soil moisture seemed to be associated with increased rate of nitrogen accumulation specially at the milk and dough stages (Fig. I). The derogatory effect of excess soil moisture on protein synthesis as emphasised by Stewart and Greaves (1909), Greaves and Carter (1928), Clements (1937) and Eaton *et al* (1952) among others was made evident in that crude protein of the wheat plant was inversely related to moisture of the soil supporting the crop in the present investigation.

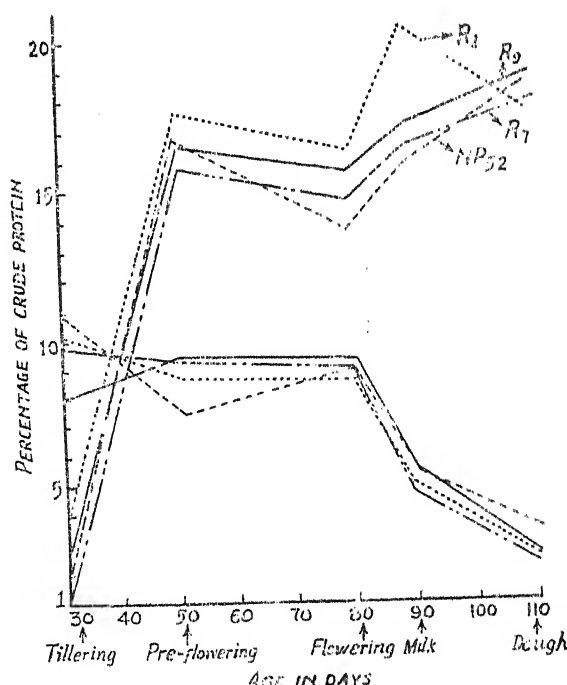


Fig. I. Relationship between crude protein of the wheat plant and moisture of the soil supporting the crop.

The crude protein status in tops of the wheat plants NP₅₂ and its X-ray progenies at their different physiological stages of growth clearly shows the time effect of X-rays for which the NP₅₂ seeds were exposed. Except R₇ in the tillering and R₁ and R₇ in the dough stages, the crude protein status of the wheat tops was

comparatively high at all the stages in the X-ray strains. The figures amply show increased protein metabolism in the plant tops possibly arising from X-ray treatment at least upto the milk stage. However, in the inflorescence in the later stages of

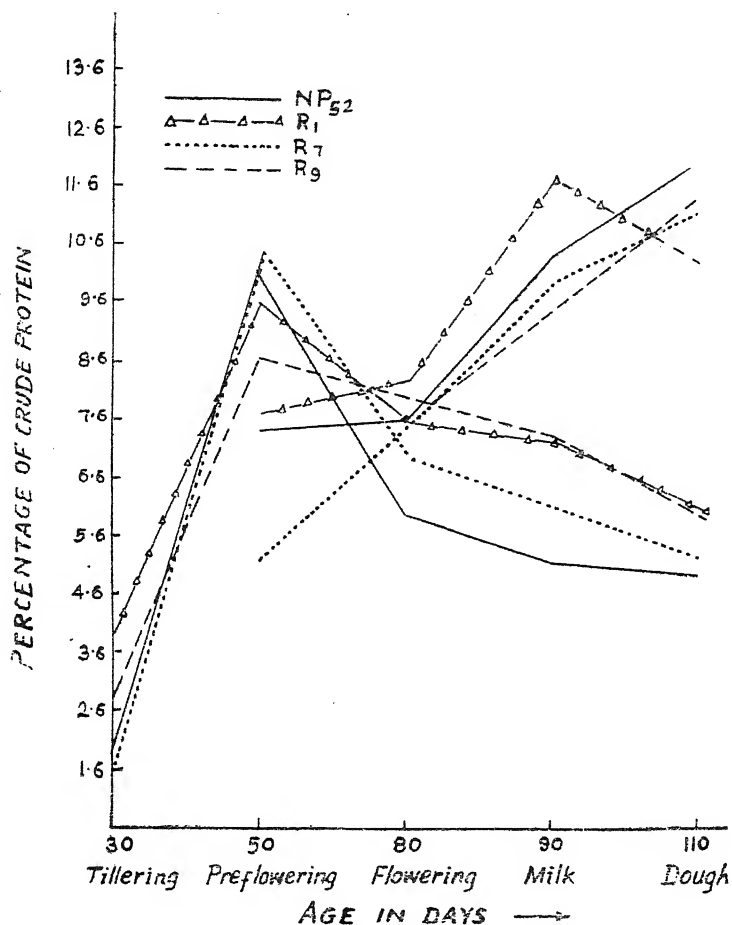


Fig. II. Distribution of crude protein between the vegetative and the reproductive stages of the wheat plant.

growth especially in the dough, the decreased amount of crude protein in the X-ray progenies shows its derogatory effect upon protein accumulation in the seeds.

TABLE III. Number of tillers of wheat strains at successive stages in the life-cycle

(Average, per plant)

| Physiological stages (days) | Strains | Tiller Number |
|--------------------------------|------------------|---------------|
| Tillering (30) | NP ₅₂ | 7.963 |
| | R ₁ | 5.107 |
| | R ₇ | 5.242 |
| | R ₉ | 5.106 |
| Preflowering (50) | NP ₅₂ | 9.178 |
| | R ₁ | 8.345 |
| | R ₇ | 7.796 |
| | R ₉ | 7.899 |
| Flowering (80) | NP ₅₂ | 8.928 |
| | R ₁ | 7.999 |
| | R ₇ | 7.714 |
| | R ₉ | 7.785 |
| Milk (90) | NP ₅₂ | 8.856 |
| | R ₁ | 8.213 |
| | R ₇ | 7.714 |
| | R ₉ | 7.641 |
| Dough (110) | NP ₅₂ | 8.819 |
| | R ₁ | 7.996 |
| | R ₇ | 7.714 |
| | R ₉ | 7.636 |

From the results it is evident that the adverse effects of X-rays on protein metabolism in wheat plants are manifested at their later stages of growth while in the earlier stages there is increased protein metabolism. Whether this low value of crude protein of the inflorescences in the X-ray strains in relation to their parental plant NP₅₂ is due to its increased transformation into other essential

substances like carbohydrates and fats, is yet under investigation. It is indeed an important point to note that wheat plants arising from the X-ray treated seeds produce in the long run seeds of low protein value which is definitely a matter of importance since protein is considered to be one of the essential foods products for all living beings.

TABLE IV. Number of green leaves of wheat strains at successive stages in the life-cycle

(Average, per plant)

| Physiological stages (days) | Strains | Green leaf number |
|--------------------------------|------------------|-------------------|
| Tillering (30) | NP ₅₂ | 23·035 |
| | R ₁ | 25·606 |
| | R ₇ | 25·857 |
| | R ₉ | 27·463 |
| Preflowering (50) | NP ₅₂ | 37·314 |
| | R ₁ | 35·171 |
| | R ₇ | 36·353 |
| | R ₉ | 37·003 |
| Flowering (80) | NP ₅₂ | 36·071 |
| | R ₁ | 34·353 |
| | R ₇ | 35·396 |
| | R ₉ | 36·856 |
| Milk (90) | NP ₅₂ | 25·396 |
| | R ₁ | 24·321 |
| | R ₇ | 24·856 |
| | R ₉ | 25·642 |
| Dough (110) | NP ₅₂ | 7·880 |
| | R ₁ | 7·470 |
| | R ₇ | 7·609 |
| | R ₉ | 7·330 |

TABLE V. Negative relationship of grain yield (per acre) with the percentage difference of crude protein between the preflowering and the flowering stages of growth of the X-ray strains.

| | X-ray strains | | |
|--|----------------|----------------|----------------|
| | R ₁ | R ₇ | R ₉ |
| Average yeild of grains per acre (in lbs.) | 862·8 | 1340·8 | 1651·1 |
| Percentage difference of crude protein between the preflowering and the flowering stages | 1·3375 | 1·1250 | 0·5000 |

SUMMARY AND CONCLUSION

Wheat seed NP₅₂ and its three X-ray progenies R₁, R₇ and R₉ were the material for study.

These wheat strains viz., NP₅₂, R₁, R₇, and R₉ were grown under field conditions in randomized blocks in sandy loam soils of the Indo-Gangetic Plains at Allahabad and the total nitrogen of tops of the plants was evaluated at the tillering, preflowering, flowering, milk and dough stages.

Translocation of nitrogenous food material from the vegetative tops to the developing inflorescences was noted from the preflowering stages onwards. Decline in the number of green leaves and the tillers was noticed to be positively correlated with the total nitrogen of plants at flowering. Reduction in field moisture had an appreciable effect on the crude protein content of the wheat plants at milk and dough stages. Yield of X-ray strains was found to be influenced by the fall of crude protein at flowering; this fall was negatively correlated with the yield of strains. Wheat plants arising from X-ray treated seeds produced seeds of low protein value.

The status of nitrogen and crude protein and their transference to the different parts of the plant were found to depend on the X-ray treatment, variety, physiological stages of growth, number of green leaves and tillers and also soil moisture conditions.

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STUDIES INTO THE GROWTH BEHAVIOUR AND YIELD OF NP WHEAT AND ITS X-RAY PROGENIES.

By

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INTRODUCTION

In a previous communication (1960) it was shown that the X-ray progenies of NP₅₂ behaved differently than the latter. It was thought fit to examine the variance induced if any, in the germination of NP₅₂ and its X-ray progenies and the relationship of the % of germination to the growth behaviour of the plant as judged by relative stand of plants, vertical growth, tillering capacity (tiller number) and dry matter accumulation, and also the final yield in the two cases.

Wheat being an important cereal was chosen for this series of investigations. The additional advantage was that the dormancy factor was eliminated as wheat seeds are known to germinate even without a rest period since with dormant seed the evaluation effect of X-ray may not be so clear cut.

It is not uncommon to think that the growth behaviour and yield of plants are controlled, at least partly, by the germination percentage of their mother seeds. While sufficient data are lacking as to the effects of X-rays upon this factor, other investigations relating to the general derogatory influence of X-rays upon plants' life processes may well be visualized to have the indential results on the germination behaviour of wheat seeds. Johnson (1936), however, recorded the opinions of a few workers that small doses of such radiations might sometimes exhibit stimulatory effects upon the growth behaviour as well as other aspects of plants in general.

The present work was undertaken to study the variations in germination percentage of NP₅₂ wheat and its three X-ray mutants as well as the complex relation that this factor had with growth viz., acre-stand of plants, number of tillers, height, dry matter accumulation and yield.

EXPERIMENTAL

Germination Percentage. The percentage of germination of wheat seed of different strains viz., NP₅₂, R₁¹, R₇² and R₉³ were determined in the laboratory. Randomly selected seeds in lots of hundred were placed over filter papers kept in petridishes of suitable size to each of which 2 c. c. distilled water was added daily at 10 a. m. To safeguard the seeds from all probable sources of damage the petri dishes were kept under a glass germination chamber. The number of seeds germinated were noted each day at 10 a. m. The germinated seeds were removed daily while the ungerminated seeds were retained in the petridishes for further observations that continued until no more germination took place. The germinated seeds of the strains were kept in other petridishes and allowed to germinate. The vigour of the seeds was judged fourteen days after germination (Plate I).

1, 2 and 3: R₁, R₇ and R₉.

These Ranjan strains were evolved by X-ray treatment of New Pusa₅₂ (NP₅₂) seedlings for varying periods (Ranjan, 1940).

Some strains of seed were sown in the Agricultural farm of the Department in replicated randomized manner and their growth behaviour was studied. Seven areas of one yard square were marked at random in each plot after sowing had been done.

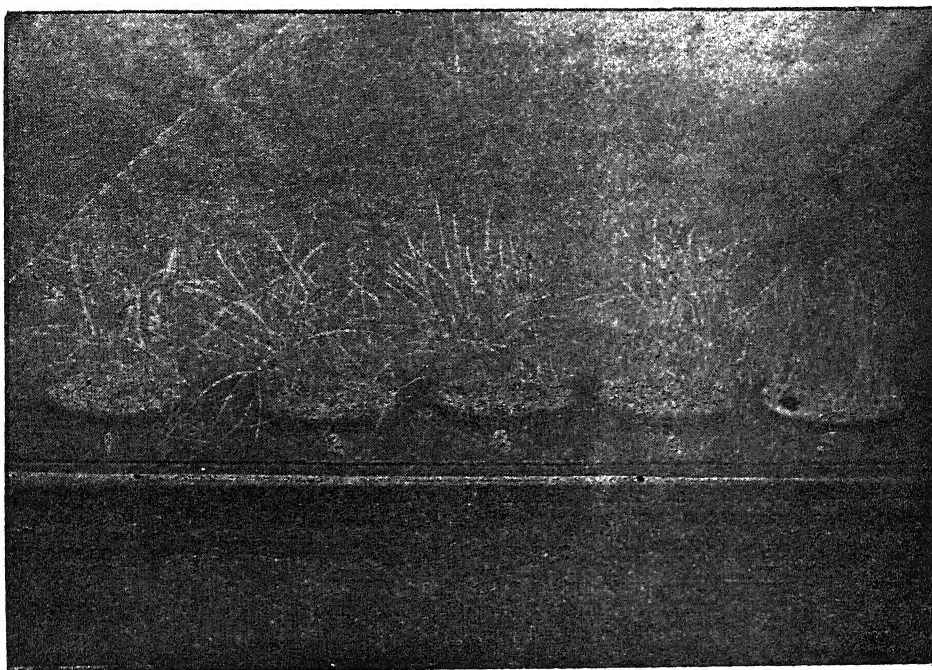


Plate I. Vigour of the seedlings fourteen days after germination ;

1. C₁₃; 2. NP₅₂; 3. R₁; 4. R₇; 5. R₉.

As the seedlings attained about 4" height one plant in each quadrat was demarcated for the morphological records. Dry matter accumulation data were collected on plants uprooted from outside the demarcated one yard-square areas, so that the competition among the neighbouring plants remained unaffected throughout the period of study.

Relative stand of plants, vertical growth, tillering capacity (tiller number), dry matter accumulation, yield of grain, straw and husk for each strain was recorded at the physiological stages (tillering, preflowering, flowering, milk and dough) in the life-cycle of the plant.

The quadrat was taken as the unit for all quantitative measurements and the values were computed to an acre-basis.

EXPERIMENTAL FINDINGS

Germination Percentage. The X-ray mutants seemed to be better than their parental plant New Pusa 52 in germination percentage (Table I). However, a perusal of the germination percentage of the seeds in the first 24 hours showed

clearly the slow rate of germination of the X-ray progenies R_1 and R_7 as against NP_{52} , the control.

Table I. Percentage of germination of wheat seeds.

(No. cent)

Temp. $25^{\circ}\text{C} \pm 1$

| Strains | Time in hours | | Total |
|-----------|---------------|----|-------|
| | 24 | 48 | |
| NP_{52} | 40 | 55 | 95 |
| R_1 | 25 | 73 | 98 |
| R_7 | 8 | 92 | 100 |
| R_9 | 64 | 31 | 95 |

Relative Stand. NP_{52} seedlings recorded the maximum stand (Table II) in comparison with X-ray progenies in the following order:

$$NP_{52} > R_9 > R_1 > R_7$$

Table II. Relative stand of plants of different wheat strains at tillering.

(per acre)

| Strains | Number of plants |
|-----------|------------------|
| NP_{52} | 2,54,975 |
| R_1 | 2,33,575 |
| R_7 | 2,22,275 |
| R_9 | 2,52,565 |

Tillering. Johnson (1939) found increased tillering in working with certain wheat seedlings from soaked grains treated with 1000 r-units and 5000 r-units than did controls. In the present investigations, however, no such stimulation was found among the r-strains, instead their parental plant recorded maximum tiller numbers at all the physiological stages of growth while as regards this factor the X-ray mutants

interchanged their positions among themselves until the milk stage after which they recorded their stabilised ranks (Table III).

Table III. Variations in the behaviour of plants of wheat strains at successive stages in the life-cycle.

| (Average/ plant) | | | | |
|-----------------------------|------------------|---------------|-----------------------------|-----------------------------------|
| Physiological stages (days) | Strains | Tiller number | Vertical growth (in inches) | Dry matter Accumulation (in gms.) |
| Tillering (30) | NP ₅₂ | 7.963 | 9.692 | 1.719 |
| | R ₁ | 5.107 | 8.967 | 1.863 |
| | R ₇ | 5.242 | 9.449 | 1.474 |
| | R ₉ | 5.106 | 11.667 | 1.796 |
| Preflowering (50) | NP ₅₂ | 9.173 | 34.924 | 8.015 |
| | R ₁ | 8.345 | 27.767 | 5.888 |
| | R ₇ | 7.796 | 29.406 | 6.626 |
| | R ₉ | 7.899 | 31.454 | 7.207 |
| Flowering (80) | NP ₅₂ | 8.928 | 36.235 | 9.280 |
| | R ₁ | 7.999 | 28.203 | 8.020 |
| | R ₇ | 7.714 | 33.428 | 7.960 |
| | R ₉ | 7.785 | 31.565 | 8.460 |
| Milk (90) | NP ₅₂ | 8.856 | 36.546 | 10.600 |
| | R ₁ | 8.213 | 28.178 | 9.460 |
| | R ₇ | 7.714 | 30.871 | 9.470 |
| | R ₉ | 7.641 | 31.744 | 9.970 |
| Dough (110) | NP ₅₂ | 8.819 | 36.703 | 10.420 |
| | R ₁ | 7.996 | 28.357 | 9.360 |
| | R ₇ | 7.714 | 30.912 | 8.655 |
| | R ₉ | 7.636 | 31.763 | 9.710 |

Vertical Growth. Inspite of its maximum record of vertical growth in the tillering stage, the position of R_9 became next to NP_{52} from the pre-flowering stage onwards when the parental plant recorded the maximum vertical growth (Table III).

Dry Matter Accumulation. But for the tillering stage, NP_{52} recorded the maximum dry matter accumulation from pre-flowering stage onwards. All the three R-strains accumulated dry matter in much less quantity than NP_{52} after the tillering stage (Table III).

Grain Yield. Except R_1 , both R_9 and R_7 recorded larger yield of grains as compared to NP_{52} , R_7 being immediately next to R_9 in its position of highest yield measure (Table IV).

Table IV. Average yield of grain, straw and husk
Yield, per acre

| Strains | Grain | | | Straw | | | Husk | | |
|-----------|-------|-----|---------|-------|-----|---------|------|-----|---------|
| | Md. | Sr. | Ch. | Md. | Sr. | Ch. | Md. | Sr. | Ch. |
| NP_{52} | 14 | — | 35 — 3 | 28 | — | 35 — 0 | 5 | — | 31 — 14 |
| R_1 | 10 | — | 20 — 15 | 33 | — | 10 — 3 | 5 | — | 36 — 4 |
| R_7 | 16 | — | 14 — 1 | 28 | — | 35 — 0 | 6 | — | 26 — 14 |
| R_9 | 20 | — | 5 — 7 | 33 | — | 14 — 12 | 7 | — | 28 — 7 |

Md.=82 lbs. approx.

Yield of Straw and Husk. All the R-strains recorded maximum yields of straw and husk in comparison with their parental plant NP_{52} (Table IV).

DISCUSSION

The maximum germination recorded by R_9 in the first 24 hours may be due to the time effect to which the NP_{52} seedlings were exposed to X-rays giving rise to these R_9 mutants. In the second 24 hours bulk of germination of R_1 and R_7 took positions. It may be noted that the NP_{52} seeds recorded, under both the time durations practically the same percentage of germination, the difference between the percentages of germination in both the first and the second 24 hours being not so large as the X-ray mutants. This unequal germination of the X-ray progenies clearly showed the physiological unbalance in the metabolic processes of the seeds at the time of their germination brought about by X-ray treatment. Wort (1941) recorded a few investigations which also reported deleterious effects of X-ray radiations upon plant growth.

A photograph of the germinated seedlings in petridishes was taken after 14 days of germination in order to note the vigour of these growing seedlings (Plate

I). It seemed that R_9 was the best, better than NP_{52} which was, however, not the case, because the rate of growth of R_9 as regards height was not found to be equal to NP_{52} which even after surpassing the growth in height in the petridishes stood erect and unlodged. Among the X-ray strains the stand of R_1 and R_7 seemed to be weaker than NP_{52} as practically no differential growth between them could be traced out.

While the effect of a more balanced germination of NP_{52} in the first and the second 24 hours is evident upon the tiller number of these plants, a vigorous and rapid gremination of R_9 in the first 24 hours seemed to have affected their number of tillers in the long run. With the progress of growth period, R_1 and R_7 held their respective intermediate positions parallel to their positions in germination percentage in the first 24 hours. Total germination percentage of NP_{52} and its X-ray progenies seemed to have no influence on their habit of tillering.

Record of low stand by the X-ray progenies in comparison with their parental plant NP_{52} was in accordance with Johnson (1939) who also with certain wheat seedlings from soaked grains treated with 1000 r-units and 5000 r-units showed less growth in all respects (except increased tillering) than controls. Low acre stand of the R-strains only showed the minimising effects of X-ray radiations upon their stand.

It is evident (Table I) that the difference in germination percentage between the first and the second 24 hours had sufficiently influenced the relative stand of the wheat strains under consideration. The relative stand seemed to be inversely related with this difference of germination percentage. The effect of X-rays in increasing the magnitude of this difference had indirectly affected the relative stand of the R-strains. No relationship could be traced out between the total germination capacity and relative stand of NP_{52} and its X-ray progenies.

The depressing effects of X-ray treatments seemed to emerge in the long run among the R-strains which recorded low vertical growth from pre-flowering stage onwards.

A perusal of Table I would show a fair inverse relationship between the difference of percentage germination in the first and the second 24 hours and the vertical growth of the strains taken for this study. In this case also there seemed to no relationship between the total germination percentage and the vertical growth of the strains under consideration.

The deleterious effects of X-ray treatments became evident in the later stages among the R-strains which recorded low dry matter accumulation than NP_{52} . Francis (1934) also in working with wheat seedlings treated with X-rays noted retardation of dry weight of the growing parts of these young plants. With the progress of the growing period a good reverse relationship became evident between the difference of germination percentage in the first and the second 24 hours and the accumulation of dry matter among NP_{52} and its X-ray progenies.

While there seemed to be no relationship present between the grain yield of the strains and their percentage of germination, maximum yield of grains recorded by R_9 might be conjectured to have been influenced by its highest record of germination in the first 24 hours. Low record of grain yield by R_1 seemed to be more due to the time-effect of X-rays to which the NP_{52} seedlings were exposed giving rise to these R_1 strains. Highest yield of straw and husk recorded by the X-ray progenies

seemed to be due to the stimulating effects of X-ray treatments upon the vegetative parts of the plants as Shull and Mitchell (1933) noted in some behaviour of wheat stimulative action as a result of using metallic screens, high voltage, low amperage and brief exposures (approx. 100 r-units) of such crops to X-rays. It might be pointed out, however, that the stimulative action resulting to the high yields of straw and husk of R-strains might only be due to the time-effect of X-rays to which the NP₅₂ seedlings were exposed and not due to any technical difference as was shown by Shull and Mitchell (1939) which was avoided in the present investigations. In addition to the time-effect of X-ray treatment, the maximum yield of straw and husk recorded by R₀ strains might also be due to its better germination in the first 24 hours. However, no clear relationship could be established between the straw and husk yield and germination percentage.

SUMMARY

An attempt to study the percentages of germination of a few X-ray strains and their parental seed NP₅₂ was made under laboratory conditions and some explanation put forward to make clear the complex relationship between this factor and other behaviours of the plant i.e., stand, vertical growth, tillering, dry matter accumulation and yields of grain, straw and husk. The plants were grown in the Gangetic sandy-loam soil of Allahabad.

While NP₅₂ recorded a balanced germination percentage in both the first and the second 24 hours, all the X-ray strains showed better germination in the second 24 hours. R-strains recorded higher percentage of total germination than NP₅₂.

Total germination percentage seemed to have little influence upon the morphological behaviour, dry matter accumulation and yield. In most cases, the percentage of germination in the first 24 hours and the difference of germination percentage between the first and the second 24 hours to have more influence upon the different growth behaviours and yield of the strains.

Higher yields of grains, straw and husk of R-strains were explained to be mostly due to the stimulative effect of X-ray treatment as well as the time-effect to which the NP₅₂ seedlings were exposed giving rise to these R-strains.

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